Bt Cotton





Questions and Answers

K. R. Kranthi



K. R. Kranthi

Ph.D, FNAAS Director Central Institute for Cotton Research, Nagpur

Published by INDIAN SOCIETY FOR COTTON IMPROVEMENT (ISCI), MUMBAI



@Indian Society for Cotton Improvement, 2012 All rights reserved. No part of this publication may be reproduced in any form or by any means, electronically, mechanically, or by photocopying, recording or otherwise, without the prior permission from the author or owners of the copyright.

Publisher: The Secretary Indian Society for Cotton Improvement, (ISCI) c/o Central Institute for Research on Cotton Technology Adenwala Road, Matunga, Mumbai 400 019

Price: Rs. 200/- (postage extra)

Printer: Surya Offset Printers, Nagpur

Foreword

'No question is so difficult to answer as that to which the answer is obvious'-George Bernard Shaw.

The success story of Bt-cotton in India is obvious, but it has indeed become strangely, circumspect to affirmatively answer the 'obviously easy to answer' question -'has Bt-cotton succeeded in India in combating the bollworm menace?' The answer lies in the simple fact that farmers have endorsed the technology in a vast majority. If Bt-cotton would not have controlled bollworms, the technology would not have moved the distance it has today.

There may be a need for refinement and constant changes are always inbuilt into science. While we progress with advanced technologies for sustainable growth and prosperity, environment should always be uppermost in our minds. Questions must be asked and concerns will be raised, but, science must provide answers and solutions. Bio-safety concerns are paramount to all of us. Answers should be forthcoming from good robust scientific experiments. We need not shy away from moving forward to develop GM technologies in a manner that is profoundly acceptable to the ecology, environment and society. But, any new technology must be compared to the previously used technologies and evaluated for the trade-off benefits, checks and balances and economic gain of the farmers.

It is clear that there is hardly any technology that can be 100.0% safe to everything. Interestingly, Bt-cotton is one of the few technologies having the safest bio-safety profiles. It comes as an alternative to the previously used hazardous concoction of insecticide mixtures. The insecticides used on cotton were known to have ravaged ecology, disrupted the environment, played havoc with human and animal health, were toxic to honey bees, insect-parasitoids and predators, caused allergies and a myriad number of ill-effects. Bt-cotton removed that to a great extent. Strangely, this seems to have been less acknowledged by detractors of the Bt-cotton technology. It is true that insecticides are now being used for sap-sucking pest control on Bt cotton hybrids, but, as mentioned in this book, the increase is because of the susceptible hybrids and has nothing to do with Bt-technology. We cannot afford to move back towards the pesticide era. By all scientific standards, Bt GM Cotton technology is by far the most environment friendly technology available thus far. We must however develop varieties and hybrids that show comprehensive resistance to sucking pests through resistant germplasm sources and to bollworms through Bt genes. This is possible through good plant breeding efforts. Once this is done, it is for sure that insecticide usage will be substantially reduced.

Bt-cotton was the first of GM technologies to be introduced into India. It is beyond doubt that farmers preferred Bt-cotton instead of the hazardous insecticide-cocktails for bollworm control. It is true that because of huge investment potential, multinational companies had the edge to develop the technology more efficiently and at a faster pace, compared to many public sector institutions across the world. But, GM technologies are being developed now more easily than before, as the transformation technology itself has advanced tremendously. India cannot afford to lose the competitive edge in agriculture, in the international arena, by slowing down biotechnology applications in agriculture. While we move forward, it surely becomes everybody's responsibility to use the best science based technologies available to the farmer after weighing out all concerns and consequences, but, we need to move forward to face future challenges of burgeoning food and clothing demands of the ever-increasing populace.

I congratulate Dr Kranthi for the good effort in bringing out all possible facets of the Bt-cotton technology, especially from the Indian perspective, in the form of questions and answers, which makes the book readable. I hope that this book will enable all stakeholders for better understanding so as to assist in proper assessment of the technology in as rationally a manner as possible.

Prof S. K. Datta Deputy Director General (CS). ICAR

Preface

A prudent question is one-half of wisdom - Francis Bacon

This book has hundred questions on Bt-cotton. These are representative of the many questions that I have been asked either by farmers, scientist colleagues, parliamentarians, family and friends. I tried to put across my views, as a cotton scientist who has been in the thick of Bt-cotton, since its inception. I believe in what I wrote in the form of answers. There could be many more questions, for which answers are not easy. One of them is, why do we need 1128 hybrids and more to come, in India? Honestly, the reply is –'I don't know'.

What do we make of Bt-cotton, after 10 years of its tumultuous and tremendous journey in India? Did it succeed in what it was supposed to do? Clearly, the answer is -'yes, Bt cotton was expected to control only bollworms and it succeeded in doing just that, protecting cotton crop against bollworms all through the 10 years'. Yields may have increased because of effective protection against bollworm damage and also because of many other technologies and factors that were simultaneously introduced. But, most importantly, insecticide usage for bollworm control decreased by more than 90.0% As an entomologist who has seen bollworm larvae refusing to die even with the strongest possible concentration of pyrethoids and insecticide-cocktails, I know the value of Bt-cotton. I do not hesitate to express my gratitude to the technology, because it came in at a time when farmers needed it the most.

What are the major criticisms? There are views that 'Bt-cotton is unsuitable for rainfed regions', 'insecticide usage has increased with Bt-cotton', 'input usage has increased with Bt-cotton', 'India's yields stagnated irrespective of the increase in Bt-cotton area', I tried to answer all these in an unbiased manner. But basically, these issues have nothing to do with Bt-technology. These are related to the -sucking-pest susceptible Bt-hybrids needing more insecticide, most of which are long duration, are unsuitable for rainfed regions, cause water and nutrient wastage with excessive foliage and many a times do not perform well in marginal soils of rainfed regions.

Criticism also focused on 'biosafety issues ranging from adverse impact on soil microbes, goats, sheep, cattle, and reported presence of Bt in human blood and placenta'. These reports were characterized by methodology errors and clearly lacked scientific credence in establishing a clear 'cause and effect relationship'.

Thus, I believe that the criticisms in India on performance are related to hybrids and have less to do with the Bt-technology per-se. Bt-technology was supposed to control bollworms, and it did splendidly. It was commercial considerations and market forces that did not enable harnessing full potential of the technology in India. What could have been different at the policy level for India to have been able to harness the full potential of the Bt-technology? I believe that, along with the Bt-hybrids, presence of Bt-varieties could have changed the Indian cotton scenario. Also, while GEAC and RCGM should have focused only on biosafety approval, instead of evaluating and approving the 1128 Bt-hybrids, the identification of appropriate Bt-hybrids or varieties should have been the domain of ICAR (Indian Council of Agricultural Research) and the NARS (National Agricultural Research) system. This would have made a great deal of difference to the Indian cotton scenario.

Dr Manjunath wrote a very useful book with 85 questions on Bt-cotton in India. Interestingly, majority of the questions in this book are different, from what are presented in his book. I am sure that there may be more questions. But, to take away credit from a good technology such as Bt-cotton, not crediting it with what it deserves, is only possible if the information and understanding are incomplete. This book provides basic information and data to enhance understanding on several issues. There are tables of data in this book, which have been personally compiled by me and I will stand by them.

In all things that we do, if some can influence some influential somebody resulting in betterment of farmers and the environment, some of us would have indeed done something. This book is a humble attempt in this direction. Happy reading -Keep questioning.

-K. R. Kranthi



I thank Dr C. D. Mayee, President ISCI for kindly consenting to publish this book and for the tremendous support always. I thank Professor S. K. Datta, DDG (CS), ICAR, for the kind foreword and the guidance and encouragement. I thank Dr S. Ayyappan, Secretary DARE and DG, ICAR, for his kind support.

I would like to profusely thank Dr G. V. Ranga Rao, (formerly with Bayer, India), Mr Anurag Bhatnagar, Sudarshan Chemicals, Mr Sanga Reddy, Indofil, and A. R. Reddy, Senior Scientist, CICR for their help and assistance with insecticide usage data and compilation. I thank Dr Sandhya Kranthi, Head Crop Protection, Dr Blaise Desouza, Head, Crop Production and Dr Venugopalan, Principal Scientist, CICR for editing, helpful discussions and suggestions with this book. Mrs Mukta Chakrabarty and Dr M. S. Yadav, deserve sincere thanks and appreciation for their efforts in producing this book. I thank Mr Sampath Kumar for the patient proof reading. Dr Chinna Babu Naik and Mr Sabesh, scientists of CICR and Dr Udikeri of UAS Dharwad, provided the photographs. I thank them.

I can't name the farmers, but would like to thank them immensely for having asked questions, which are now part of this book.

Finally, I thank my colleague and wife Dr Sandhya for being with me all through thick and thin, many a times only to suffer my incorrigible idiosyncrasies. My mother Mrs Savithri provides inspiration for everything that I do. And, now my 12 year old daughter Divya has joined the list of persons who inspire me to work more. My heartfelt gratitude to them.



Contents

Chapter 1: INTRODUCTION

- 1. What is Bt-cotton?
- 2. Why do we need Bt-cotton?
- 3. Do we need other Bt-crops?
- 4. How does Bt-cotton kill insects?
- 5. Is Bt-cotton really selectively toxic to insects?

Chapter 2: BIOSAFETY

- 6. Is Bt-cotton toxic to goats and cattle?
- 7. Is Bt-cotton harmful to human beings?
- 8. Are the current bio-safety-testing methods adequate?
- 9. Does Bt-cotton enter the food chain?
- 10. Is GM food crop acceptance low in other countries?
- 11. Does Bt-cotton impact biodiversity through pollen-flow?
- 12. Is the native Desi cotton under threat because of Bt-cotton?
- 13. What are terminator seeds- are the terminator genes used anywhere?
- 14. Does Bt transformation have negative effects on the physiology of the plant?
- 15. Does Bt-cotton disrupt ecology and environment?
- 16. What are the biosafety tests conducted in India and abroad?
- 17. Who can conduct biosafety tests in India?
- 18. Who examines the bio-safety test results for approval of the Bt-cotton hybrids?

Chapter 3: BT TECHNOLOGY IN INDIA

- 19. Who developed the technology?
- 20. Which are the Bt genes available in Bt-cotton commercialized in India?
- 21. How many Bt-hybrids are available in India?
- 22. Has the cotton area increased after the introduction of Bt-cotton?
- 23. What has been the trend of Bt cotton uptake in the various states?
- 24. What were the major changes in production in various states of India?
- 25. Which states made the biggest strides in productivity or lint yields per hectare?

6

26. How many countries are cultivating Bt cotton?

Chapter 4: YIELDS AND PRODUCTION ASPECTS

- 27. Have yields increased in India after the introduction of Bt-cotton?
- 28. Are the yields stagnating in India and why?
- 29. Can the average yields increase in India now above the average of 500 kg/ha?
- 30. Has the cost of cultivation increased because of Bt cotton?
- 31. Is it possible to reduce the cost of cotton cultivation and enhance yields?
- 32. Is the increase in yield because of Bt cotton alone?
- 33. What are the current major production constraints that can be overcome to enhance yields?
- 34. Have the yields increased globally due to Bt-cotton?

Chapter 5: HYBRIDS AND WATER REQUIREMENT

- 35. Is Bt-cotton best suited only for irrigated regions?
- 36. If Bt-cotton is best suited to irrigated conditions and better soils, then, why is it that the yield in North India are only at 500-600 kg/ha which is less than half of USA, Australia and Brazil?
- 37. Is Bt cotton suitable for rainfed conditions, especially in Vidarbha of Maharashtra?
- 38. Are farmers unhappy with Bt-cotton in India?
- 39. Are farmer suicides related to Bt-cotton in Vidarbha?
- 40. Has Bt-cotton benefitted Vidarbha?

Chapter 6: BOLLWORM CONTROL

- 41. Is Bt cotton really controlling bollworms?
- 42. How many sprays are being used now to control bollworms in cotton?
- 43. Have there been changes in insecticide usage for bollworm and sucking pest control?
- 44. Is the pesticide use increasing again now and why?
- 45. Have bollworm populations declined because of Bt cotton?
- 46. Do the bollworms find new host crops because of Bt cotton?
- 47. Have the bollworm populations increased in non-Bt crops?
- 48. Do bollworms still occur on Bt cotton?
- 49. How can bollworms still feed on Bt cotton?
- 50. Is the toxin expression variable in hybrids, plant parts and over the season?

Chapter 7: RESISTANCE AND MANAGEMENT

- 51. What is refugia?
- 52. Are farmers following the refugia guidelines?
- 53. Will bollworms develop resistance to Bt-cotton?
- 54. What are the various resistance management strategies being adopted across the globe?
- 55. How is bollworm resistance monitored?
- 56. Have bollworms developed resistance to Bt-cotton so far?
- 57. Has the pink bollworm developed resistance to Bt-cotton in India?
- 58. What kind of approaches can be adopted to delay resistance?
- 59. Do we have 'India-specific' IRM strategies to delay resistance?

Chapter 8: NEW GM COTTON TECHNOLOGIES

- 60. Are there new Bt genes for new versions of Bt-cotton?
- 61. Are we going to continue with addition of new genes for insect control?
- 62. What is herbicide resistant cotton?
- 63. Can herbicide resistant cotton be grown with inter-crops?

Chapter 9: BENEFITS OF BT COTTON

- 64. What have been the benefits of Bt-cotton so far in India?
- 65. Has there been any environmental benefit with Bt-cotton?
- 66. How can we improve the environmental benefits?
- 67. What have been the benefits of Bt-cotton, so far, abroad?
- 68. Has the quality of cotton improved with Bt-cotton?
- 69. Did our imports decline and exports increase with Bt-cotton introduction?

Chapter 10: CRITICISMS AND CONCERNS

- 70. What has been negative fallout of the cultivation of Bt cotton on the farmers field?
- 71. What are the major criticisms of Bt-cotton by NGOs and concerns expressed by farmers?
- 72. Is parawilt associated with Bt-cotton?
- 73. What are the perceived risks with Bt-cotton?
- 74. Are new pests and diseases appearing on cotton and why?
- 75. Why is it that the cotton leafworm *Spodoptera litura* is able to feed on Btcotton?
- 76. How are these new diseases and insect pests related to Bt-cotton?
- 77. How much of Bt toxin gets accumulated in the soil?

- 78. Have the soils become depleted of nutrients because of Bt-cotton?
- 79. Are the Cry toxins harmful to soil insects?
- 80. Has Bt-cotton adversely affected soil microbes and soil health?

Chapter 11: HYBRIDS v/s VARIETIES

- 81. How can we harness the full benefits of Bt-cotton in India?
- 82. Why do we have Bt-cotton only as hybrids in India?
- 83. Which countries other than India cultivate hybrid cotton?
- 84. Do the new GM technologies have potential to increase yields in India?
- 85. What is the advantage of straight varieties over Bt-hybrids?

Chapter 12: BT and ORGANIC COTTON

86. Is Bt-cotton compatible with organic cotton?

- 87. Does Bt-cotton contaminate organic cotton?
- 88. Are there alternate technologies apart from Bt cotton for pest management?

Chapter 13: ILLEGAL BRANDS AND SPURIOUS SEEDS

- 89. Can the farmers detect Bt-cotton seeds or plants?
- 90. What is the status of spurious Bt-cotton hybrid seeds in the market?
- 91. What are illegal, fake and spurious seeds?

Chapter 14: POLICY PERSPECTIVES

- 92. Are there problems with IPR?
- 93. Can Bt-cotton contribute to food-security?
- 94. What is the status of public sector cotton in India?
- 95. What kind of policies will help in enhancing cotton yield in India?
- 96. Which are the policies and IPM strategies that may augment cotton pest management?
- 97. Are there any specific policy issues for cost effective profitable cotton cultivation in rainfed regions?
- 98. Is GM labeling necessary?
- 99. Will India lose competitive edge if we slow-down on GM technology?
- 100. Can India become a world leader in cotton?



1. What is Bt-cotton?

Bt cotton is genetically modified cotton crop that expresses an insecticidal protein whose gene has been derived from a soil bacterium called *Bacillus thuringiensis*, commonly referred as Bt. Many subspecies of *B.thuringiensis* are found in soils and are in general known to be toxic to various genera of insects but safe to other living organisms. Bt was first discovered by a Japanese scientist Ishiwata in the year 1901. Bt has been used as an insecticide for control of stored grain pests since 1938 in France and from 1961 as a registered pesticide in the USA and later in many other countries including India as sprays in cotton IPM programs to control insects. Bt toxins thus have several decades of proven selective toxicity to insect pests and with established safety record to non-target animals. Currently there are 67 recognized subspecies of *B. thuringiensis* most of which produce spores and insecticidal proteins.



The *B.thuringiensis* strains produce three types of insecticidal toxins, crystal (Cry) toxins, cytolytic (Cyt) toxins and vegetatively expressed insecticidal proteins (vip). These toxins are highly specific to certain insect species. Thus far until September 2012, a total of 229 cry toxins (Cry1Aa to Cry72Aa), 11 cyt toxins (cyt1Aa to cyt3Aa) and 102 vip toxins (vip1Aa1 to vip4Aa1) have been discovered. A total number of 342 Bt toxin genes are available for research to develop insect resistant GM crops.

The Bt gene *cry1Ac* was used to develop the first Bt-cotton variety. The gene was transferred into the genome of cotton explants (tissue pieces) using a bacterium called *Agrobacterium tumefasciens*. The transformed cells were developed into a full GM plant now called Bt-cotton. In general, Cry1Ac toxins are highly specific to insects at species level, and are not known to cause any harm to non-target species such as fish, birds, farm animals and human beings.

Currently, Cry1Ac, Cry2Ab and Cry1C have been approved for commercial cultivation in India. Bt cotton hybrids available in India are derived from technologies developed by Monsanto (Cry1Ac and Cry1Ac + Cry2Ab), Metahelix (Cry1C), Chinese Academy of Agricultural Sciences through Nath seeds (modified Cry1Ac called as fusion gene) and JK seeds (Cry1Ac). Dow Agrosciences are conducting field trials with Cry1Ac + Cry1F and Bayer is introducing Cry1Ab + Cry2Ae. There were 1128 Bt-cotton hybrids in 2012, developed by 40 seed companies, available in the Indian markets.

2. Why do we need Bt-cotton?

- a) Cotton is a long duration crop and is attacked by large number of insect pests throughout its growth and development.
- b) The three bollworms, American bollworm *Helicoverpa armigera*, Pink bollworm *Pectinophora gossypiella* and the Spotted bollworms, *Earias vittella* and *Earias insulana* are major pests and cause serious threat to cotton production resulting in significant yield losses.



Helicoverpa

Pectinophora

Earias

- About 9400 M tonnes of insecticides worth Rs 747 crores were used only for bollworm control in 2001
- d) Before the introduction of Bt cotton, insecticide quantity applied on cotton was the highest, relative to other cultivated crops.
- e) Cotton bolls are highly vulnerable to hidden insects such as the American bollworm, pink bollworm and spotted bollworm.
- f) Bollworms, especially the pink and spotted bollworms are hidden feeders and generally do not come into direct contact with insecticide sprays.



- g) The American bollworm which comes into contact with insecticides, partially, has developed resistance to almost all the insecticides recommended for its control in all regions of the world.
- h) Nearly 90.0% of all insecticides in Pakistan and about 50.0% of all insecticides in India were being unsuccessfully used for cotton pest control, until the year 2001, before Bt cotton was introduced. Of these insecticides about 70.0% was for bollworm control and the rest for sap-sucking insects.
- i) Resistant sources are unavailable in the germplasm and resistance breeding has been unsuccessful.

3. Do we need other Bt-crops?

Transgenic/GM technology holds promise for the benefit of mankind as it allows precision breeding. GM technology should be the method of choice only when conventional methods fails to deliver. Prioritization of crops and traits should be of prime importance. In today's context, emergence of new pests and diseases and global climate change are the major challenges that need to be addressed on an urgent basis to meet demands of growing population. Limited availability of genetic variability makes it difficult to utilize conventional breeding as the only method of choice. Hence, conventional breeding in combination with GM technology will be an option that needs to be employed in order to maintain self sufficiency in food production as well as to bring an evergreen revolution in our country.

Three crops, paddy, cotton and pigeon-pea are major consumers of insecticides in India. Insecticides worth Rs 4215 crores were used for insect pest management in agriculture in India in 2010, out of which Rs 1250 crores (30% of the total) were used on paddy, Rs 880 crores (21%) were used on cotton and Rs 332 crores (8%) were used on pigeon pea in 2010 in India. Other crops such as chilles consumed insecticides worth Rs 231 crores (5.5%), soybean Rs 154 crores (3.7%), Bengal gram Rs 146 crores (3.5%) and Brinjal Rs 146 crores (3.1%). Despite the use of insecticides, crop losses due to insect pests are estimated at 30 to 50% in these crops because of cryptic pests such as bollworms, pod borers, stem borers and fruit and shoot borers that are well protected from external pesticide application and require highly hazardous systemic insecticides that are absorbed by plant tissues.

The use of Cry toxins to develop GM crops in paddy, pigeonpea, chillies and soybean has the potential to reduce the use of hazardous insecticides on food crops. Thus far there have been no credible scientific reports of Cry1Ab or Cry1Ac toxicity to human beings, despite the Bt food crop, Bt-maize, being consumed directly for about a decade.

However, if Bt crops of pigeonpea, chillies, chickpea, tomato, soybean etc., are developed using the Cry toxins that are in use currently, these may add to the selection pressure to enhance resistance development in the American bollworm *H. armigera* to Cry toxins. Therefore it would be appropriate to make an informed choice of genes to be used for pest management in alternate host crops for *H. armigera* management so as to ensure that there is no cross-resistance with the existing toxins.

4. How does Bt-cotton kill insects?

The Cry1Ac, Cry2Ab, Cry1C, Cry1F etc., belong to the class 'Bt-delta-endotoxins' which function as oral toxins. The delta-endotoxins are ingested and the protoxins present in the crystals are proteolytically activated to trypsin-resistant active core δ -endotoxin in the alkaline mid-gut. The active toxin traverses the peritrophic membrane to bind cadherin receptors present on the brush border membrane of the insect midgut. The cadherins process the toxins to form homo-oligomers and bind to specific receptors like alkaline phosphatases and aminopeptidases before causing pores in the epithelial membrane, resulting in osmotic lysis of the cells. This results in cessation of feeding and finally causing mortality.

Amongst the genes that have been deployed in insect resistant transgenic cotton, thus far, Cry1Ac is the most toxic to *H. armigera* and against a wide range of lepidopteran insect pests that include the other two bollworms the spotted bollworm *Earias vittella* & the pink bollworm, *Pectinophora gossypiella*. Bt-cotton incorporated with Cry1Ac is highly toxic to the bollworms and other minor pests such as the cotton semilooper and hairy caterpillar, but not effective on the leaf eating tobacco caterpillar *Spodoptera litura*. Cry2Ab2 (present in Bollgard-II) is moderately toxic to *Helicoverpa armigera*, and *Spodoptera litura*. However the expression levels of Cry2Ab2 are very high (120-350 ppm), as compared to the Cry1Ac (0.5 to 15 ppm) and therefore Bollgard II is highly toxic to *H. armigera* and toxic to *Spodoptera litura*.

The Cry1F is not effective on *H. armigera* but the levels of expression in 'Widestrike' (Dow Agrosciences) is at 10-40 ppm that confers mild tolerance to *H. armigera* and fairly good toxicity to *Spodoptera litura*.

5. Is Bt-cotton really selectively toxic to insects?

The Cry toxins are specifically toxic to specific classes of insects. For example the Cry1Ac is toxic to three species of cotton bollworms, but is less toxic to the tobacco caterpillar, *Spodoptera litura* and is non-toxic to other classes of insects which are sap-sucking pests such as mealybugs, jassids, aphids, whiteflies etc. Other Cry toxins as Cry1F and Cry1C are more toxic to tobacco caterpillar *Spodoptera litura* and relatively less toxic to the cotton bollworms.

The Cry1Ac is mainly toxic to the bollworms (cotton bollworm, pink bollworm and spotted bollworm), semiloopers and hairy caterpillars. Bt-cotton expressing Cry1Ac is non-toxic to other non-target organisms such as beneficial insects, birds, fish, animals and human beings. Laboratory and field studies carried out in India showed that the Cry1Ac protein deployed in Bt-cotton did not have any direct effect on any of the non-target beneficial insects. Work carried out elsewhere in the world also showed similar results.

Hilbeck et al., (2012) confirmed their earlier findings that Cry1Ab toxin increases mortality of the two-spotted ladybird beetle, *Adalia bipunctata* larvae. The results point towards the possibility that a few toxins may have an enlarged spectrum of toxicity spanning across insect orders such as Lepidoptera and Coleoptera.



6. Is Bt-cotton toxic to goats and cattle?

Cry toxins have not been reported to be toxic to higher animals such as goats, sheep and cattle in any part of the world. However, it is only in India that apprehensions were expressed by NGOs regarding sheep mortality at Warangal and Adilabad district of Andhra Pradesh due to grazing in Bt cotton fields. The issue was examined by the State Government and reports received from the Directorate of Animal Husbandry, Hyderabad and the Indian Veterinary Research Institute, Izatnagar, U.P. revealed that the sheep deaths might be due to high content of Nitrates/Nitrites, residues of hydrocyanide (HCN) and organophosphates which are common constituents of pesticides used during cotton cultivation and not due to Bt toxin.

Scientific evidence indicates that the possibility of Cry toxins killing goats and sheep is remote. The Cry toxins do not get activated under the acidic conditions of nontarget animals such as goat, sheep and cattle. Feeding studies did not show any toxicity symptoms that could lead towards extreme toxicity symptoms or mortality.



Comprehensive biosafety studies were carried out by ICAR institutions with Bt cotton. First the safety of Bt Cry protein on lab animals such as rabbit, rat and guinea pigs. Various studies such as primary skin irritation test on rabbit, irritation to mucous membrane in rabbits, acute oral toxicity study in rats and skin sensitization study on guinea pigs were conducted. The results showed that Bt protein and Bt-Cotton seed powder were non-irritant to the skin of rabbits and vaginal mucus membrane. In case of acute oral toxicity study in rats, Bt cotton seed material did not induce any treatment related observable toxic effects when compared with Non-Bt cottonseeds. Studies on skin sensitization revealed that the repeated application of Bt cottonseed extract did not induce dermal sensitization (allergies) to the skin of any of the guinea pigs when compared with animals applied with extract of non-Bt cottonseeds.

Secondly, broiler chickens were tested by feeding of Bt cotton seed meal. This study was conducted at ICAR's Central Avian Research Institute, Izatnagar. Methodical studies were conducted with broiler chickens and tested for the effect of Bt protein. Birds were weighed at weekly intervals to observe weight loss or gain. After the 7th week of study, 8 birds per treatment were sacrificed to study the effect of feeding CSM types on different carcass traits and development of digestive and immune organs. The results of the study revealed that the body weight gain and feed conversion efficiency, did not differ statistically over all phases of study. The protein and energy efficiencies of experimental diets fed to broiler chicken also remained statistically similar. The carcass traits (% of live weight) of broilers (blood loss, feather loss, dressed yield, eviscerated giblet, ready to cook yield and abdominal fat), cut up parts (breast, drum stick, thigh, back, neck, wings) and digestive and immune organs weights (heart, liver, gizzard, spleen, bursa) also remained statistically (P<0.05) similar to control. It was concluded that the solvent extracted Bt cottonseed meal can be included safely with maize or soybean meal based broiler diet up to 0-7 weeks of ade.

A systematic study was conducted with Bt cotton seed meal as a feed for Fish Common Carp and the side effects if any were tested in the fish food chain. This study was conducted at CIFE, Mumbai. A 60-day feeding trial was conducted on common carp fry. Bt cotton seed cake was included in the diet of common carp at 3 -level (10, 20, 30%) and compared with its non-Bt counterpart along with control group comprising of no cotton seed cake. Growth rate of Bt cotton seed cake fed group was comparable (P< 0.05) with that of control group and which and non-Bt counterpart as well. No mortality was found after feeding the Bt cotton cake, suggesting no adverse effect of Bt cotton seed cake.

Studies were also conducted on large animals such as Cow and Sheep to assess the bio-safety of Bt cottonseed. A trial was conducted at Central Sheep & Wool Research Institute (ICAR), Avikanagar for 120 days by continuous feeding on Weaner lambs at a higher plane of nutrition. Nutrient (OM, CP and fiber fractions) and mineral (Ca, P. Mn, Co and Zn) contents were identical in Bt-cotton and non-Bt cotton seeds. The growth performance of lambs was similar on control, non- Bt cotton seed and BTcotton seed included diets. The growing lambs consumed 168 g Bt-cotton seed per day and did not have apparent adverse effect on dry matter intake, nutrient utilization and nitrogen balance. Similarly Bt-cotton seed intake of 0.681 % of body weight or 19.5 % of dry matter intake did not produce deleterious effect on performance and dry matter intake, thus palatability and growth performance was not a problem for Btcotton seed feeding in lambs even under high plane of nutrition. Rumen fermentation characteristics viz, pH, TVFA and NH3-N concentrations was not influenced by feeding of GNC, non- Bt cotton seed or Bt-cotton seed in lamb diets, Heamatological observations did not change due to Bt-cotton seed feeding compared to non-BT cottonseed or GNC feeding. Interestingly feeding of Bt-cotton seed increased RBC and decreased WBC in blood. Serum IgG level did not change due to Bt and non-Bt cotton seed feeding. Thus feeding of BT cottonseed to lambs did not alter immunity and allergen status. Internal organs weights as g per kg empty live weight (ELW) indicated precise effect of Bt- cottonseed feeding on internal organ changes. The weights of kidney, spleen, pancrease, heart, lung, penis, kidney fat, cole fat, GI tract, ingest and empty GI tract were not different among Bt cotton seed and non-Bt cotton

seed fed lambs. However, Bt cotton seed feeding increased liver weight, testicle weight and testicle fat g/kg empty live weight. The results were considered to indicate no detrimental effects.



A comprehensive study was conducted with Bt cotton seed meal on milking cows. This study was conducted at NDRI, Karnal for four weeks. Sixteen crossbred (KS and KF) multiparous cows were adapted to test by feeding Bt cottonseed based diet. Mainly the Bt Cry protein side effect and absorption in the milk was tested. Milk yield and voluntary feed intake were recorded daily while milk samples were collected at the start of experimental feeding and thereafter at weekly intervals during the four week experimental period for the analysis of milk composition and to test for the presence of Bt protein. At the end, a blood sample from each cow was collected and plasma was separated to test for the presence of Cry 1Ac protein. Cry 1Ac protein in cottonseed, milk and blood samples was measured by ELISA method. The amount of Cry 1Ac protein in Bt cottonseed was 195.04 ng/g on fresh basis. Corresponding value in Bt concentrate mixture was 78 ng/g on fresh basis. Cows in both the groups improved their body weight during the study period and body weight gain in both groups was similar. Average milk yield during 28 days of experimental period in Non Bt (13.53 kg/day) and Bt (13.12 kg/day) groups did not vary significantly. During the experimental period the milk composition in terms of fat, protein, lactose, SNF and total solids content in Bt and Non-Bt were similar. Cry 1Ac protein was not detected in milk samples, drawn at 0, 7, 14, 21 and 28 day of feeding the experimental diet, as well as in plasma samples drawn on day 28 from the cows fed the Bt cottonseed

based ration. Lactating dairy cows of both the groups did not show symptoms of any disease, maintained their health and performed in a similar fashion when fed with Non Bt and Bt cottonseed as a source of energy and protein supplement during the fourweek long experimental period. The present study results revealed that the Cry1Ac proteins were neither detected in the milk nor in blood of cows that were fed with Bt cottonseed during the four week trial. Further, there was no effect of Bt cottonseed containing Cry protein on milking cows. Hence, feeding of Bt cottonseed as a source of protein and energy in the ration of crossbred cows was considered to be safe and as nutritious as Non Bt cottonseed.

A field study was carried out at CICR, Nagpur by a team of scientists led by a senior scientist of the Krishi Vigyan Kendra, for two years (2007-2009) by tethering six goats in one hectare of Bt cotton and one hectare of conventional cotton. The goats were fed on the crop continuously for four months and there were no differences in any biological aspects of the two sets of animals. The biochemical and health results clearly showed that Bt cotton was safe to goats.

7. Is Bt-cotton harmful to human beings?

The Cry toxins Cry1Ac, Cry1Ab, Cry2Ab, Cry1F and Cry1C are considered to be safe to human beings. The stomach of humans, being the first organ of digestion the Bt protein encounters, is acidic and contains proteases like pepsin which degrade the Bt protein. Thus the alkaline conditions needed for pro-toxin solubilization and protease action required for toxin activation are absent in the stomach. More importantly the human intestine lacks the specific receptors to which the activated Bt protein binds and initiates the physiological effect. Bt-cotton is being cultivated in at least 12 countries and was cultivated in at least five major countries for more than a decade. Cotton seed oil is a by-product of Bt-cotton and is used in all the cotton growing countries. But, Bt-toxins or Bt-DNA were not detected in refined oil.

Bt maize was first cultivated in the year 1996 in USA followed by Canada, European union, South Africa, Argentina, Honduras, Philippines, Uruguay, Czech republic, Chile, Romania, Brazil and Egypt. To date there are 16 countries which are growing GM maize on commercial scale. In 2010, USA was leading the countries by occupying 317 lakh ha followed by Brazil (73 lakh ha), Argentina (30 lakh ha), South Africa (19 lakh ha) and Canada (12 lakh ha) and remaining 11 countries such as Uruguay, Philippines, Spain, Chile, Honduras, Portugal, Czech Republic, Poland, Egypt, Slovakia, Romania occupied less than 10 lakh ha. The area under Bt maize in 2010 was 102 lakh ha.

Farmers in South Africa grow two types of maize varieties namely Yellow and White maize. Yellow maize is raised by commercial farmers for animal feed, cornstarch, and corn syrup. White maize with Bt was cultivated in 11.4 lakh ha in South Africa for human consumption. Bt maize is being cultivated for more than 10 years without any harmful effect on human beings and other non-target organisms. China approved the commercial cultivation of Bt-Rice (genetically modified with *cry1Ac* gene) from 2009. Though several NGO organizations raised bio-safety issues from time to time, these were found to have in-sufficient credence, especially in light of the extremely high

hazards posed by insecticides when used as an alternative to the GM based pestcontrol technology.

The two main concerns debated have been on possible "allergenicity" of GM food and "gene transfer" of the transgene into human cells. The FAO Food and Agriculture Organization of the United Nations and the World Health Organization (WHO) evaluated protocols for tests for GM foods. No allergic effects were found in GM foods currently on the market.

Gene transfer. The WHO states that "Gene transfer from GM foods to cells of the body or to bacteria in the gastrointestinal tract would cause concern if the transferred genetic material adversely affects human health. This would be particularly relevant if antibiotic resistance genes, used in creating GMOs, were to be transferred. Although the probability of transfer is low, the use of technology without antibiotic resistance genes has been encouraged by a recent FAO/WHO expert panel". Netherwood et al., (2004) conducted experiments by feeding GM soybean to human volunteers and found that the transgene did not survive passage through the gastrointestinal tract.

Aris et. al., (2011) reported that 93 per cent of blood samples taken from pregnant women and 80 per cent from umbilical cords tested positive for the presence of Cry1Ab from Bt-corn in both non-pregnant women and pregnant women and their fetuses. The paper questioned the assumption that Cry toxins do not enter human blood stream. Subsequently several authors (De Wech, Marcel Kuntz and organizations such as Food Standard Australia New Zealand) (FSANZ) found the paper unconvincing and were critical of the validity of ELISA technique to detect Cry1Ab in human blood and that too at concentration that was less than detection limits of ELISA of 1 ng/ml. The values of 0.04 to 0.19 ng Cry1Ab per ml of blood serum or tissue, are below the detection limits of 1 ng/ml of the AgDia ELISA kits used. Though no adverse effects were reported by the authors, because of the reported presence of Cry1Ab in human blood, if true, the information may have significance to long term biosafety concerns that will have to be examined. Therefore, the results should have been confirmed through real-time PCR using event-specific primers for MON810 of Bt-corn. This was neither carried out by Aris et al., 2011 in the paper, nor was it done subsequently by any other laboratory. Considering the tremendous impact the report had, it was expected that several independent laboratories would reconfirm the findings. However, the work has not as yet been supported by any further evidence either from the same lab or any other research teams after it was first reported in 2011.

8. Are the current bio-safety-testing methods adequate?

In a recent review (Snell et al., 2012) published in 2012, a team of scientists from UK and France reviewed 12 long-term studies (of more than 90 days, up to 2 years in duration) and 12 multigenerational studies (from 2 to 5 generations). They concluded that "Results from all the 24 studies do not suggest any health hazards and, in general, there were no statistically significant differences within parameters observed. The studies reviewed present evidence to show that GM plants are nutritionally equivalent to their non-GM counterparts and can be safely used in food and feed"

In a report on GMOs published in 2010

(http://ec.europa.eu/research/biosociety/pdf/a_decade_of_eu-funded_gmoresearch.pdf),

the European Commission Directorate-General for Research and Innovation state that "The main conclusion to be drawn from the efforts of more than 130 research projects, covering a period of more than 25 years of research, and involving more than 500 independent research groups, is that biotechnology, and in particular GMOs, are not per se more risky than e.g. conventional plant breeding technologies. Another very important conclusion is that today's biotechnological research and applications are much more diverse than they were 25 years ago, which is also reflected by the current 7th EU Framework Programme".

Key et al (2008) reviewed GM food biosafety and state that "Foods derived from GM crops have been consumed by hundreds of millions of people across the world for more than 15 years, with no reported ill effects (or legal cases related to human health) despite many of the consumers coming from that most litigious of countries, the USA. There is little documented evidence that GM crops are potentially toxic"

However, organizations such as Greenpeace still contend that the current biosafety testing methods are inadequate. Séralini et al., (2007) scrutinized the industry-funded biosafety data on rats tested with rootworm resistant (MON863) maize and pointed out differences in kidney size and blood composition. Le Curieux-Belfond *et al.* (2009) reanalyzed the industry-funded biosafety data and concluded that three rat feeding safety data for glyphosate resistant maize, root-worm resistant maize and borer resistant maize actually showed liver, kidney, and heart damage in the rats. Séralini et al (2011) reviewed 19 studies of mammals fed with commercialized genetically modified soybean and maize which represent, per trait and plant, more than 80% of all environmental genetically modified organisms (GMOs) cultivated on a large scale, after they were modified to tolerate or produce a pesticide. They concluded that "*The 90-day-long tests are insufficient to evaluate chronic toxicity, and the signs highlighted in the kidneys and livers could be the onset of chronic diseases. However, no minimal length for the tests is yet obligatory for any of the GMOs cultivated on a large scale, and this is socially unacceptable in terms of consumer health protection.."*

The European Food Safety Authority (EFSA) re-examined the criticism by Seralini of the safety data and concluded that the observed small numerical decrease in rat kidney weights were not biologically meaningful, and the weights were well within the normal range of kidney weights for control animals. The EFSA questioned the statistical methods used by Seralini and pointed out that the methods were wrong and the interpretation by Seralini unacceptable. Finally, the EFSA, The French High Council of Biotechnologies Scientific Committee (HCB) and the Food Standards Australia New Zealand were highly critical of the Seralini reports only to conclude that the three GM maize events presented no admissible scientific element likely to ascribe any haematological, hepatic or renal toxicity.

Seralini et al., (2012) reported more tumors in rats fed for 2 years with Rounduptolerant genetically modified maize cultivated with or without Roundup, and Roundup alone (from 0.1 ppb in water) as compared to control. The paper has been severely criticized for faulty statistical analysis and for using the albino Sprague-Dawley strain which is prone to mammary tumors when food intake is not restricted and also has a strong natural tendency to develop cancers.

9. Does Bt-cotton enter the food chain?

The possible routes of Bt-cotton protein entering the food chain are, through human consumption of un-refined cottonseed oil, in which traces of Bt protein may be present with particulate seed residues or through consumption of meat or milk of the animals which fed on Bt cotton seed-cake. However, ELISA tests showed that milk and meat were found to be free of Cry proteins. Thus the chances of Bt proteins entering the human food chain through milk and meat are low.

10. Is GM food crop acceptance low in other countries?

Low acceptance of GM foods is expressed only by countries in Europe. Almost all insecticide discoveries and the subsequent commercialization through multinationals based in Europe were from European countries. The multinationals in these countries and their economy depends on insecticide markets all across the globe. Food crops constitute a large share of pesticide usage and GM food crops have the potential to reduce pesticide usage significantly. While food in these countries is not a limitation, GM crops do not matter much for them, considering low food demand, less population and high purchasing power. Major NGOs such as Green peace have also their origins in Europe.

Most countries (including Europe) growing transgenic crops or importing transgenic food or feed have a regulatory system in place. These approaches are also influenced by Substantial Equivalence, Principle of Familiarity and Generally Regarded as Safe (GRAS) as working principles as well as by multilateral negotiations related to environmental and human health safety (e.g., Cartagena Protocol on Bio-safety, International Plant Protection Convention, Codex Alimentarius) and trade (e.g., Agreement on the Application of Sanitary and Phytosanitary Measures, Agreement on Technical Barriers of Trade, Agreement on Trade-related Aspects of Intellectual Property Rights) and United Nations Convention on Biological Diversity.

11. Does Bt-cotton impact biodiversity through pollen-flow?

Cotton pollen is heavy and cross pollination happens mostly through pollinator insects such as honey bees and pollen beetles. Pollen-flow from Bt-cotton can contaminate non-Bt cotton varieties if compatible for crossing. However, the cultivation of GM Bt-cotton hybrids, does not pose any risk to bio-diversity of naturally occurring Indian cotton or more specifically on the 'Western ghat biodiversity' as it was pointed out by a panel. The native biodiversity of cotton in India is represented only in the Desi-cotton species which have evolutionary origins in India and are known to have been cultivated in the country for 5000 years. Since the Desi cotton species *Gossypium arboreum* and *Gossypium herbaceum* have native origins, there is high level biodiversity of the Desi species in India. However, there is no possibility, whatsoever, of any of the native India Desi cotton species *Gossypium arboreum* and *Gossypium herbaceum* species getting genetically contaminated with GM Bt-cotton, so as to threaten the extant biodiversity. Desi cottons are diploid in their genetic constitution whereas the American cotton (*G. hirsutum*) is allo-tetraploid. Thus the Desi cottons

species and the tetraploid cotton (all Bt cotton commercialized in India are tetraploid cottons) are incompatible for cross-fertilization.



All the current Bt-cotton hybrids are of the American cotton species Gossypium hirsutum. The American cotton species Gossypium hirsutum was introduced into India in 1790 by the British East India Company and does not have much of naturally evolved biodiversity in the country. The seeds of all Gossypium hirsutum varieties that were developed in the country represent biodiversity of the American cotton species Gossypium hirsutum available in India. These are conserved and preserved in their pure form at NBPGR (National Bureau for Plant Genetic Resources) and CICR (Central Institute for Cotton Research) and can be retrieved as and when required.

12. Is the native Desi cotton under threat because of Bt-cotton?

The species *Gossypium hirsutum* is a tertraploid with chromosome number of 4n=52, and is genetically incompatible with the Desi species which are diploids with chromosome number of 2n=26. The diploid species are not crossable with tetraploid species and thus reproductively incompatible with the tetraploid species *Gossypium hirsutum*. Further, there is no record of occurrence of any tetraploid wild cotton species in India or any other Malvaceous species in India or more specifically in the Western ghats, that are crossable or even remotely likely to be contaminated with the American cotton species *Gossypium hirsutum* of Bt cotton. Many countries have restricted the option of developing GM crops in land races and native cultivars especially if the crop has evolutionary origins in the country so as to prevent erosion of biodiversity.

13. What are terminator seeds- are the terminator genes used anywhere?

Terminator technology is not permitted for use in any part of the world. The terminator technology or genetic use restriction technology (GURT) results in harvesting of inviable seeds. This necessitates farmers to buy seeds for each sowing hence increasing the chances of monopoly by MNCs. However, entry of this technology has been clearly restricted in India through PPV&FRA act- where it has been clearly mentioned that every application for registration under section 14 should be accompanied by an affidavit sworn by the applicant that such variety does not contain any gene or gene sequence involving terminator technology. Definitely, terminator seed sales would have added much larger impact on Indian farmers, as it forces farmers to purchase fresh seeds for every crop season. But, the efforts of Government of India through PPV &FRA Act avoided this negative impact on the Indian farmers

14. Does Bt transformation have negative effects on the physiology of the plant?

Commercialization of GM crops involves isolation of gene and plant transformation, selection of best events, bio safety assessment, limited scale and large scale field trials. The impact of genetic modification on physiology of the plant depends on the site of transgene integration. This is mainly because current methods of plant transformation result in the random integration of the gene of interest in the target genome. Physiology of the plant is affected only when the transgene integrates within the gene whose function is necessary for normal growth and development of crop plant. Since the integration is a random phenomenon, generation of large number of events and screening for the best event with normal physiology is an important step in GM crop development. As a bio-safety regulatory compliance, substantial equivalence studies are done through composition analysis (Protein, carbohydrate, oil, calories, ash, nitrogen, crude fibers and moisture contents) between GM and Non-GM counterpart of a crop to negate the concerns related to impact on physiology of plant.

The current Bt-cotton events available in the market do not show any adverse physiological effects and behave as normally as any conventional hybrid. In some regions the Bt-hybrids have been found to have shallow roots, due to early onset of reproductive phase. This may have also been due to hard-pan of the soils or surface irrigation during early seedling stage.

15. Does Bt-cotton disrupt ecology and environment?

Lu et al., (2012) showed that in the last 13 years GM crops delivered significant environmental benefits by reducing the insecticide usage by 50% and doubling the level of ladybirds, lacewings and spiders. Moreover, the study also stated that the environmental benefits extended to neighboring crops of maize, peanuts and soybeans.

Udikeri (2006), UAS, Dharwad, studied the dynamics of cotton aphids and predators in RCH-2Bt and non-Bt cotton hybrids. Laboratory feeding experiments using Bt and non Bt cotton were carried out to study the effect of Bt fed aphids on predator indicated no difference in incubation period, longevity of grubs and adults, fecundity and aphid consumption potential indicating safety of Cry1Ac to predator through intoxicated aphid host.

Dong et al., (2003), reported only minor effects on some life table parameters in laboratory feeding studies with lacewings and predatory beetles and none with predatory bugs and spiders. There was some evidence of a reduction in numbers of predators and parasitoids which specialise on the Bt controlled bollworms, but also of increases in numbers and diversity of generalist predators such as spiders. A decrease in the parasitoid and predator populations can be associated with decrease in the densities of the pest populations on account of Bt-cotton. Unsprayed Bt cotton sustained 4 times more attack of tarnished bugs, 2.4 times more with boll weevil, 2.8 times more with stink bugs and *Spodoptera*.



Chrysoperla adult

Ladybird beetle

Ladybird grub

Due to these changes in pest complex, farmers had to spray 3-5 times on Bollgard as compared to 6-8 times on non-Bt cottons. Any effects could be assigned to the decrease in prey quality – for example with stunted *Spodoptera litura* caterpillars which had fed on Bt cotton. There was no increase in aphid or whitefly numbers on Bt cotton. In general, such adverse effects as have been measured are very small when compared with the effects of the spraying of conventional insecticides. Insecticide usage for bollworm control has reduced by about 90.0%. The reduced use of insecticides for bollworm control has resulted in ecological and environmental benefits. However, the use of susceptible hybrids as carriers of Bt- technology in India has resulted in increasing of insecticide usage for sap-sucking pest control.

16. What are the biosafety tests conducted in India and abroad?

A series of protocols have been formulated as pre-requisite for environmental release of genetically engineered (GE) plants in India. The bio-safety tests conducted in India and abroad are almost identical. These protocols include

- 1. Acute Oral Safety Limit Study in rats or mice
- 2. Subchronic Feeding Study in rodents
- 3. Protein Thermal Stability
- 4. Pepsin Digestibility Assay
- 5. Livestock Feeding Study

These protocols address key elements of the safety assessment of foods and/or livestock feeds that may be derived from GE crops. The protocols are based on international best practices, including guidance and peer reviewed publications available from the Codex Alimentarius Commission, the Food and Agriculture Organization, the World Health Organization, the Organization for Economic Cooperation and Development, and the International Life Sciences Institute.

Tier 1 studies: These studies are conducted on non-target organisms in the laboratory using test diets incorporating concentrations of the target protein at, or above, the maximum estimated environmental exposure.

Some representative non-target organisms for tier-1 testing are, mouse, Avian model, Freshwater fish, Aquatic invertebrate, Non-target arthropods: Honey bee larvae and adults (*Apis mellifera*), Lady beetles, Green lacewing (*Chrysoperla spp.*), Parasitic hymenopteran (*Brachymeria intermedia*), Collembola and Earthworm (Lumbricus terrestris). Subsequently, livestock feeding trials as required on a case-by case basis as per the DBT publication "Protocols for Food and Feed Safety Assessment of GE Crops.

17. Who can conduct biosafety tests in India?

Bio-safety tests can be conducted by private testing labs, contract research organizations and national institutions accepted by regulatory agencies. The private testing labs and CROs are either accredited by National Accreditation Board for Laboratories or GLP Compliance Committee under DST.

18. Who examines the bio-safety test results for approval of the Btcotton hybrids?

India's regulatory system comprises of a three-tier mechanism.

Institutional Bio-Safety Committee (IBSC) comprises of expert members and functions within the institution of technology developers. The IBSC periodically discusses and ratifies in-house proposals for GM experiments to be conducted within approved laboratories and contained greenhouses

Review Committee for Genetic Manipulation (RCGM) comprises of 13 expert members with the Advisor DBT as member secretary, constituted by the Department of Biotechnology, Ministry of Science and Technology. The RCGM is empowered to approve (or not approve) applications for all *small scale research activities* in India designed to generate information on transgenic organisms. The RCGM also approves applications for experiments involving import of transgenic material (tissue, DNA, seeds, any other plant parts), limited field trials, bio-safety and toxicity studies. The role of RCGM extends to monitor the safety related aspects in respect of ongoing r-DNA projects & activities involving Genetically Engineered Organisms/Hazardous organisms and controlled field experiments of transgenic crops through the MEC (Monitoring and Evaluation Committee). The RCGM was actively involved in clearing and guiding public and private institutions in the development of transgenic crops and RDNA therapeutics

Genetic Engineering Appraisal Committee (GEAC) is a statutory body under the Ministry of environment and Forests (MoEF) The GEAC is the lead inter-ministerial body empowered to shape - by consensus - the Government's final disposition toward large-scale use and environmental release of GM organisms. The GEAC is chaired by the Additional Secretary of MoEF, co-chaired by an expert nominee from DBT, and it includes representatives from DBT, and other related Ministries. The GEAC examines applications for large scale field trials and commercial approval for transgenic crops and recommends decisions.

19. Who developed the technology?

Six Bt cotton events have been approved thus far in India for commercial cultivation. There are four Bt Cotton events expressing Cry1Ac, one event with Cry1C, and one event with Cry2Ab2. The various technology developers are:

- a. Monsanto: MON531 (Cry1Ac) event Bollgard;
- b. Monsanto: Mon15985 (Cry2Ab2) event in Bollgard-II
- c. JK seeds, India: JK Event-1 (Cry1Ac);
- d. **Chinese Academy of Agricultural Sciences**, China: GFM Cry1A (Cry1Ac), introduced by Nath seeds India;
- e. **NRCPB, New Delhi and UAS Dharwad**, India: BNLA601 (Cry1Ac) event; Commercialized by CICR, Nagpur:
- f. Metahelix, India: Event 9124 (Cry1C) event

20. Which are the Bt genes available in the Bt-cotton commercialized in India?

There are four variants of *cry1Ac* genes apart from one each of the *cry2Ab* and *cry1C* genes. All the Cry1Ac genes present in the four events released in India are chimeric fusion genes.

The *Cry*1Ac gene in the Bollgard event 531 is a chimeric gene of 3534 bp size, with the first 1398 nucleotides (corresponding to the first 466 amino acids) of *Cry*1Ab gene and rest of the 1399-3534 nucleotides (corresponding to the 467-1178 amino acids) from the *Cry*1Ac gene. Except for one amino acid at 766 position, the *Cry*1Ac amino acid sequences are identical to that of the wild type *Cry*1Ac gene. The chimeric gene produces a protein that is 99.4% identical to that of the wild type *Cry*1Ac.

The Cry1Ac genes in JK and BNLA106 are chimeric fusion genes of 1842 bp with the first 1398 nucleotides (corresponding to the first 466 amino acids) of Cry1Ab gene and rest of the 453 nucleotides (corresponding to 151 amino acids at 467-671 position) from the Cry1Ac gene.

The Cry1Ac in Nath seeds is >99% identical to the Cry1Ac used in JK and BNLA106 events except that the size is smaller at 1824 bp with the first 1377 nucleotides (corresponding to the first 459 amino acids) of *Cry*1Ab gene and rest of the 453 nucleotides (corresponding to 151 amino acids at 460-664 position) from the *Cry*1Ac gene.

21. How many Bt-hybrids are available in India?

The Bt-cotton technology was first approved in 2002 by the GEAC for commercial cultivation in central and south Indian cotton–growing zones in India in the form of three hybrids (MECH-12, MECH-162, and MECH-184). Subsequently, the GEAC approved RCH-2 (Rasi seeds) in 2004, for cultivation in the central and southern zones.

In 2005, another 16 hybrids were approved. Thus, the total reached to 20 Bt hybrids, with 6 for north, 12 for central and 9 for south India, thus making available the technology for entire country. Realizing the immense potential of the technology, several Indian Seed companies rushed forward as sub-licensees of the technology to acquire the rights to incorporate the *cry1Ac* gene into their own hybrids.

By 2006, the total number of hybrids reached 62, with an additional approval of 38 more hybrids from 15 companies, which also included the commercial release of two new Cry1Ac based events, GFM-Cry1A of China and Event-1 of JK seeds.

By 2007, an estimated total of 138 Bt-hybrids were released for commercial cultivation.

By the end of July 2008, the total number of Bt-hybrids increased to 283. By August 2009 the number increased to 564 Bt-hybrids and one Bt-variety. By August 2010 the total number of Bt-hybrids increased to 809 By May 2012 there were 1128 Bt cotton hybrids available in the market.

22. Has the cotton area increased after the introduction of Bt-cotton?

Cotton area was on the decline in India because of frequent bollworm infestation and outbreaks. The area declined from an average of 87 lakh hectares upto 2001 to a meager 78 lakh hectares in 2002 and 2003. With the advent of Bt-cotton, the area increased to 121.91 lakh hectares in 2011. Thus there was an additional increase of at least 30 lakh hectares because of the introduction of Bt-cotton.

Cotton area in some states increased significantly in three states, Gujarat, AP and Maharashtra. The area in Gujarat was 16.87 lakh hectares in 2001, but, it increased to 30.23 lakh hectares by 2011. The area in Maharashtra was 29.8 lakh hectares in 2001, which increased to 40.91 lakh hectares by 2011. The area in Andhra Pradesh was only 10.0 lakh hectares in 2001, but increased to 18.8 lakh hectares by 2011.

23. What has been the trend of Bt cotton uptake in the various states?

The uptake was rapid in Punjab, Haryana, MP, AP and Maharashtra. Desi cotton, *Gossypium herbaceum* is still cultivated in about 4.0 to 5.0 lakh hectares of 'Wagad' soils or saline belt in Gujarat and an estimated 2.0 to 3.0 lakh hectares are still under illegal Bt-hybrids. Thus the area under GEAC approved Bt-cotton hybrids is about 74.0%. The uptake in Rajasthan was slow on account of the unsuitability of several Bt-hybrids because of their susceptibility to the Cotton leaf curl virus disease. Some

regions in Karnataka are still under DCH 32 and the Desi species Gossypium herbaceum.

% Area under Bt cotton in India

| | 2002 | 2003 | 2004 | 2005 | 2006 | 2007 | 2008 | 2009 | 2010 | 2011 |
|-------------|------|------|------|------|------|------|----------|------|------|------|
| Punjab | | | | 7 | 21 | 50 | 76 | 82 | 80 | 94 |
| Haryana | | | | 2 | 8 | 46 | 66 | 82 | 87 | 92 |
| Rajasthan | | | | 1 | 1 | 10 | 26 | 67 | 68 | 70 |
| Gujarat | 1 | 3 | 7 | 10 | 19 | 35 | 40 | 51 | 56 | 74 |
| Maharashtra | | 1 | 7 | 22 | 57 | 82 | 91 | 91 | 96 | 96 |
| MP | | 2 | 14 | 22 | 49 | 80 | 98 | 98 | 99 | 99 |
| AP | | 1 | 8 | 27 | 68 | 84 | 89 | 99 | 100 | 99 |
| Karnataka | 1 | 1 | 4 | 6 | 12 | 27 | 50 | 66 | 69 | 74 |
| TN | | 3 | 12 | 12 | 38 | 67 | 97 | 60 | 74 | 82 |
| | | | | | | | <u> </u> | | | ~·~~ |

Compilation: Kranthi, CICR

24. What were the major changes in production in various states of India?

The major gains in production have been mainly from Gujarat, AP and Maharashtra. Bt cotton was introduced into North India only in 2005, and the production was already higher in north zone by then.

| | 2002 | 2003 | 2004 | 2005 | 2006 | 2007 | 2008 | 2009 | 2010 | 2011 |
|----------------|------|------|------|-------|------|------|------|------|------|------|
| Punjab | 9 | 11 | 16.5 | 21 | 24 | 22 | 17.5 | 14.3 | 16 | 17 |
| Haryana | 10 | 11.5 | 15.5 | 14 | 15 | 16 | 14 | 14.8 | 14 | 16 |
| Rajasthan | 6.5 | 8.5 | 11 | 11 | 9 | 9 | 7.5 | 11 | 9 | 15 |
| Gujarat | 33 | 50 | 73 | 80 | 103 | 112 | 90 | 98 | 103 | 114 |
| Maharashtra | 33 | 31 | 52 | 46 | 50 | 62 | 62 | 63 | 82 | 82 |
| Madhya Pradesh | 17 | 19.5 | 16 | 15 | 19 | 21 | 18 | 15 | 17 | 18 |
| Andhra Pradesh | 22 | 26 | 32.5 | 30 | 36 | 46 | 53 | 52 | 53 | 55 |
| Karnataka | 6 | 4 | 8 | 7 | 6 | 8 | 9 | 9 | 10 | 14 |
| Tamilnadu | 6 | 3.5 | 5.5 | 5.5 | 5 | 5 | 5 | 5 | 5 | 5 |
| Others | 1 | 1 | 1 | 1 | 1 | 2 | 2 | 1 | 4 | 4 |
| Loose | 8.5 | 11 | 12 | 12 | 12 | 12 | 12 | 12 | 12 | 12 |
| Total | 152 | 177 | 243 | 242.5 | 280 | 315 | 290 | 295 | 325 | 353 |

Production in lakh bales (170 kg per bale)

Source: Cotton Advisory Board

25. Which states made the biggest strides in productivity or lint yields per hectare?

Bt cotton was introduced in North India in 2005 and the yields were already high in these states. However biggest gains were made in Gujarat, Maharashtra and AP.

| Productivity / lint yi | elds per hectare |
|------------------------|------------------|
|------------------------|------------------|

| | 2001 | 2002 | 2003 | 2004 | 2005 | 2006 | 2007 | 2008 | 2009 | 2010 | 2011 |
|-------------|-------------------------------|------|------|------|------|------|------|------|------|------|------|
| Punjab | 262 | 284 | 389 | 551 | 610 | 672 | 563 | 565 | 432 | 513 | 516 |
| Haryana | 153 | 287 | 372 | 424 | 350 | 481 | 528 | 522 | 511 | 484 | 450 |
| Rajasthan | 343 | 220 | 452 | 427 | 325 | 437 | 415 | 422 | 459 | 457 | 481 |
| Gujarat | 328 | 317 | 516 | 651 | 794 | 733 | 772 | 650 | 635 | 665 | 647 |
| Maharashtra | 195 | 158 | 191 | 311 | 207 | 274 | 330 | 335 | 319 | 355 | 353 |
| MP | 546 | 561 | 565 | 472 | 521 | 505 | 540 | 490 | 424 | 445 | 433 |
| AP | 454 | 418 | 557 | 469 | 543 | 630 | 690 | 644 | 628 | 505 | 504 |
| Karnataka | 201 | 216 | 228 | 261 | 247 | 270 | 337 | 375 | 458 | 312 | 434 |
| Tamil Nadu | 425 | 600 | 619 | 725 | 607 | 850 | 687 | 780 | 817 | 697 | 702 |
| Orissa | | | | | | | | | | 459 | 459 |
| Others | 142 | 321 | 333 | 250 | 215 | 239 | 224 | 405 | 315 | 756 | 739 |
| Lint Kg/ha | 308 | 302 | 399 | 470 | 472 | 521 | 554 | 524 | 503 | 517 | 492 |
| | Source: Cotton Advisory Board | | | | | | | | | | |

26. How many countries are cultivating Bt cotton?

Currently an estimated 161 lakh hectares are under Bt cotton in 13 countries. This accounts for about 48% of the total global cotton area. Insect resistant Bt-Cotton with *cry*1Ac was first released in the US, Mexico and Australia during 1996. Later it was released in China (1997), South Africa (1998), Argentina (1998), India (2002), Colombia (2002), Brazil (2005), Costa-Rica (2008), Burkina Faso (2009) and recently in Pakistan and Mynamar in 2010. India is leading in Bt cotton acreage with about 111.9 lakh hectares at an adoption rate of 91.79 per cent followed by USA, China, Pakistan and Australia and eight countries such as Argentina, Myanmar, Burkina Faso, Brazil, Mexico, Colombia, South Africa and Costa Rica occupied with less than 5.0 lakh ha.

27. Have yields increased in India after the introduction of Bt-cotton?

| | Total area lakh ha | BG | BG- II | Bt area lakh ha | Bt area % | Lakh bales | Kg/ hectare |
|------|-----------------------|-------|-----------|--------------------|--------------|---------------|----------------|
| 2001 | 87 | | | | | 158 | 308 |
| 2002 | 78 | 0.294 | | 0.294 | 0.38 | 139 | 302 |
| 2003 | 77.85 | 0.931 | | 0.931 | 1.2 | 182 | 399 |
| 2004 | 89.2 | 4.985 | | 4.985 | 5.59 | 246 | 470 |
| 2005 | 88.17 | 10.15 | | 10.148 | 11.51 | 244 | 472 |
| 2006 | 91.73 | 36.5 | 1.5 | 38 | 41.42 | 281 | 521 |
| 2007 | 94.39 | 58.74 | 4.6 | 63.34 | 67.1 | 307 | 554 |
| 2008 | 94.06 | 55.6 | 20.4 | 76 | 80.8 | 289 | 524 |
| 2009 | 103.12 | 36.8 | 48.2 | 85 | 82.43 | 305 | 503 |
| 2010 | 111.61 | 37.4 | 63.8 | 101.2 | 90.67 | 339 | 517 |
| 2011 | 121.91 | 26.5 | 85.4 | 111.9 | 91.79 | 352 | 492 |

Yields have certainly increased after the introduction of Bt cotton in India.

Compiled by Kranthi, CICR

28. Are the yields stagnating in India and why?

Yield stagnation in India at 510<u>+</u>27 kg lint per hectare over the past 7 years from 2005 to 2011 is primarily because of the vast majority of inappropriate hybrids. Bt cotton technology cannot be blamed for this. Bt protects the crop against bollworms and a few other caterpillars and does nothing else. But in India, Bt cotton is available only as Bt hybrids available in over 1,000 brands, while in the rest of the world Bt cotton is available only as a few straight varieties. India should also have had Bt technology in straight varieties.

Most of the Bt hybrids are of 180-to 200-day duration and are not suited for rain-fed conditions. Hybrid seeds are costly and are generally sown late after ensuring adequate soil moisture to avoid the economic burden of re-sowing. Late sown maturing hybrids suffer from severe moisture stress during the critical period of peak boll formation, which takes place much later after the rains recede. The moisture stress is higher in rain-fed regions in shallow, marginal soils, which do not hold water adequate to support boll formation. This results in low yields.

Hybrids also tend to be input-intensive, so they are not suitable for at least half the area in the country, which is under marginal soils in rain-fed regions. Additionally, many hybrids are susceptible to sap-sucking insects, leaf-curl virus and leaf reddening, adding to input costs. Bt cotton in India was approved in 2002. Before 2002, the area under "non-Bt" hybrid-cotton was less than two per cent in north India

and about 40 per cent in central and south India. By 2011, more than 96 per cent of the cotton area was under hybrid cotton, more specifically the Bt hybrid. For rain-fed regions, especially with shallow-marginal soils, characterized by low input use, early-maturing straight varieties are the best option. The main advantage with straight varieties is that farmers can reuse farm-saved seeds and can take the liberty of early dry sowing, even before the onset of the monsoon, without having to worry about the risks of poor germination and re-sowing.

29. Can the average yields increase in India now above the average of 500 kg/ha?

Yields in India are at 500 kg/ha, despite the fact that the best of all available technologies are currently available to the Indian farmer. Further, there are no major biological constraints of pests or diseases which are reducing the yields. The chances of development of new high yielding hybrids that can increase the yields, are also low. The yields in rain-fed regions are also less. Thus, if yields have to be doubled, it would be appropriate to work out new strategies especially for rainfed regions. There are examples of countries such as Brazil and China where yields of 40 to 50 Q/ha are obtained under rainfed conditions using high density planting of straight varieties.

30. Has the cost of cultivation increased because of Bt cotton?

The general cost of cotton cultivation has increased over the past 5 years. This increase may not necessarily be related to Bt-cotton, but could be a result of inputintensive nature of the hybrids. The Bt-cotton hybrid seed cost is Rs 930 for 450 g pack. At least four packets are required per hectare. The cost on conventional non-Bt varieties/hybrids could be several times less than the hybrids. But, the cost of insecticide usage on conventional varieties/hybrids could be high for bollworm control and may not be as efficient as the Bt-technology.

31. Is it possible to reduce the cost of cotton cultivation and enhance yields?

The cost of cultivation can be reduced by using early maturing, short duration straight varieties, in high density planting systems. This will help in reducing the cost of seed, pesticides and weed management. In view of sustainability and establishment of a resilient crop production system, varieties always score better as compared to hybrids. Hybrids are inherently responsive to high level of inputs and are profitable in high-input intensive systems. High yields can also be obtained by cultivating straight varieties with low input costs.

32. Is the increase in yield because of Bt cotton alone?

Though GM Bt cotton technology has brought down pesticide use by about 50 per cent, it is not correct to assume that cotton yields in India doubled only because of Bt cotton.

Bt cotton was introduced in 2002 primarily for bollworm control. Subsequently, there has been a significant leap in the cotton production. During 2001 India produced about 158 lakh bales, which increased to 243 lakh bales in 2004 and 345 lakh bales by 2011. However, it is interesting to note that the yield increase by 2004 was mainly due to the IPM/IRM strategies, new insecticides, new hybrids, new area in Gujarat, apart from the 5.4% area under Bt cotton. The area under non-Bt straight varieties was about 55.0% in 2004 and non-Bt hybrids at 38.0%. Cotton Advisory Board data show that cotton yields increased by about 60 per cent in three years between 2002 and 2004 when the area under Bt cotton was a meager 5.6 per cent and the area under non-Bt cotton was 94.4 per cent. The yields did not increase significantly more than the pre-Bt era even until 2011 when the Bt cotton area touched 96 per cent.



The area under irrigation increased mainly in Gujarat after the year 2000 especially in the form of check-dams in the Saurashtra belt which had new areas of about 8-9 lakh hectares under cotton. Currently about one-third of India's production is derived from the state which has one-fourth of the cotton area. Clearly, apart from the contribution of Bt cotton, the increase in yield may have also been due to other major changes in the past 8 years. Some perceptible changes include, implementation on IPM and IRM on a large scale by the Ministry of Agriculture and ICAR, the introduction of some excellent cotton hybrids, increase in cotton area in Gujarat from 15 lakh ha to 26 lakh ha, increase in check dams and drip irrigation systems, increase in hybrid cotton area from 40% to 90% and introduction of 6-7 new effective insecticide molecules for bollworm control and sucking pest management

33. What are the current major production constraints that can be overcome to enhance yields?

Almost all production constraints in India have been overcome and conditions are unlikely to get any better in the near future. Therefore possibilities of any further yield enhancement looks weak.

- a) Bollworm is no longer a problem
- b) Bt cotton and new effective insecticides are available
- c) Bt hybrids, Bunny, Mallika, RCH-2, RCH 134, MRC-6301, MRC-6304, Tulasi-4, Brahma, JKCH 1947, Ankur 2534, Ankur 651, Ajeet-11, ACH 33, KDCHH 621, ACH 5, Eswar, Jay, Kanak etc., have good fibre traits and are high yielders.
- d) The area under Bt hybrids is now about 90%
- e) Area under irrigation has reached about 44-45%
- f) Emerging problems (mealybugs, leaf reddening, wilt, etc.,) are being effectively tackled.

Despite all the above positive factors the yields appear to have been stagnating at 300 to 350 lakh bales.

The current production constraints are related to the non-availability of early maturing short-duration Bt-varieties or Bt-hybrids that are resistant to sucking pests, suitable for rainfed regions of the country.

34. Have the yields increased globally due to Bt-cotton?

The yield increases range from 29.0 to 82.0% in the four major countries which are cultivating Bt-cotton

| Country | Year of | GM | Total* | Adoption | Yield* | Yield* | % |
|-----------|---------|--------|--------|----------|---------|--------|----------|
| | release | cotton | cotton | rate | before | (2011) | increase |
| | | Area | Area | | release | | In yield |
| | | 2010 | 2010 | | | | |
| | | (Lakh | (lakh | | | | |
| | | ha) | ha) | | | | |
| India | 2002 | 94 | 111.4 | 82.45 | 292 | 531 | 82 |
| USA | 1995 | 40 | 43.3 | 92.37 | 602 | 886 | 47 |
| China | 1997 | 35 | 51.5 | 61.16 | 890 | 1326 | 49 |
| Australia | 1996 | 05 | 5.9 | 84.74 | 1425 | 1839 | 29 |

Source: International Cotton Advisory Committee, ICAC, Washington

Chapter - 5 HYBRIDS AND WATER REQUIREMENT

35. Is Bt-cotton best suited only for irrigated regions?

Bt cotton technology is in the form of only Bt-hybrids. In general, hybrids have a longer duration with large number of bolls per plant, with a long fruiting window period. The boll retention is high in Bt-cotton plants and therefore there is a need for continuous supply of soil moisture and nutrients. Such plants need more water and nutrients to ensure proper nutrition for the developing bolls all through the fruiting phase. Irrigated regions can provide such conditions and therefore ideal for high yielding Bt-cotton hybrid cultivation.

36. If Bt-cotton is best suited to irrigated conditions and better soils, then, why is it that the yields in North India are only at 500-600 kg/ha which is less than half of USA, Australia and Brazil?

It is true that the soils of Punjab, Haryana and Rajasthan are fertile and highly suited for cotton cultivation. However, the major problem with north India is the unsuitability of hybrids for the region. The high levels of soil fertility and excessive irrigation allow the hybrids to express vigour in the form of excessive vegetative growth, which does not actually translate into high yields and nutrients are wasted.



Leaf curl virus infected plants in north India

Until 2005 100% of cotton area in north was under varieties. Now almost the entire area is under hybrids in Punjab and Haryana and 70 to 80% of the area is under hybrids in Rajasthan. Productivity in north India is likely to be under stress because of the following reasons:

- a. Declining potential of hybrids
- b. Re-emergence of Leaf Curl Virus problem due to introduction of the large number of untested hybrids in North India.
- c. High level of susceptibility to sucking pests (varieties are tolerant)
- d. Problems of nutrient deficiencies and physiological disorders due to non-descript hybrids
- e. Mealybugs and miscellaneous insect problems are likely to increase

37. Is Bt cotton suitable for rainfed conditions, especially in Vidarbha of Maharashtra?

Bt-cotton technology is highly suited for all conditions including rainfed and irrigated. The unsuitability is only related hybrids, especially the long duration hybrids that suffer moisture stress at boll formation stage, due to poor water retention of shallow soils in rainfed regions. Thus, productivity of cotton in rainfed regions including Vidarbha region is low. Protective and supplemental irrigations for cotton are not possible in 97% of the area in Vidarbha which is under rain-fed conditions. Water and nutrient requirement during peak boll formation phase are most critical for high yields. Rainfall starts in June and recedes in September. Boll formation in long duration varieties and hybrids starts in October and reaches a peak in November. Boll formation and retention get negatively affected due to low soil moisture, especially in shallow soils thus resulting in low yields. Surveys showed that soils with very low moisture retention capacity have been found to produce low yields in long duration cotton hybrids.

38. Are farmers unhappy with Bt-cotton in India?



The area under Bt cotton increased to more than 92.0% of the total cotton area of 121.91 lakh hectares in 2011 in the country.

There is a huge demand characterized by long farmer queues each year for Bt-cotton seeds of specific brands.

Thus, it may be only reasonable to presume that farmers are happy with Bt-cotton in India.

In rainfed regions with marginal soils, the performance of hybrid cotton is not satisfactory. Farmers often incorrectly attribute such poor performance with Bt cotton technology.



39. Are farmer suicides related to Bt-cotton in Vidarbha?

Studies were conducted by CICR using field survey from 720 respondent samples farmers from 120 villages of 24 Tahsils in Wardha, Yeotmal and Nagpur districts in Vidarbha region during 2006-07, 2007-08 and 2008-09. During 2007-08 only 35% area was under Bt cotton in the districts which increased to 98% in 2008-09. The net profit in 2007-08 was Rs. 1855/- while in 2008-09 it was Rs. 6209/-. Again the data analysed for Wardha and Nagpur districts where almost all farmers reported cultivation of Bt cotton, the net returns among farmers was 5722/ha in Wardha and Rs. 6733/ha in Nagpur. Prior to the introduction of Bt cotton hybrids, farmers had incurred huge expenditure on insecticides for the control of bollworm and net returns realized in most cases were negative. Farmers could not get back returns on what was invested. With the introduction of Bt cotton, the environment. Thus, it is unlikely that cultivation of Bt cotton in Vidarbha region could be linked in any way to instances of farmers suicides

40. Has Bt-cotton benefitted Vidarbha?

Introduction of Bt cotton in Vidarbha has resulted in reduction of pesticide usage and increase in yields. Prior to the introduction of Bt cotton, the average seed-cotton yield was only 4 q/ha which increased to 7.48 q/ha in 2007-08 and 10 q/ha after 2007. The productivity was low at an average of 150 kg lint per hectare during 1999 to 2005. The productivity increased to an average of 290 Kg lint per hectare over the five year period 2006 to 2010.

| | Akola | Amravati | Buldhana | Chandrapur | Nagpur | Wardha | Washim | Yavatmal | Average |
|------|-------|----------|----------|------------|--------|--------|--------|----------|---------|
| 1999 | 166 | 130 | 174 | 191 | 278 | 222 | 151 | 171 | 170 |
| 2000 | 116 | 82 | 69 | 112 | 151 | 193 | 86 | 90 | 102 |
| 2001 | 143 | 119 | 153 | 120 | 163 | 154 | 153 | 119 | 135 |
| 2002 | 171 | 148 | 197 | 129 | 165 | 161 | 144 | 132 | 154 |
| 2003 | 155 | 174 | 237 | 210 | 264 | 229 | 141 | 173 | 188 |
| 2004 | 113 | 136 | 128 | 165 | 235 | 219 | 122 | 146 | 146 |
| 2005 | 118 | 148 | 196 | 171 | 229 | 186 | 177 | 148 | 160 |
| 2006 | 365 | 151 | 214 | 310 | 212 | 227 | 172 | 202 | 224 |
| 2007 | 334 | 342 | 379 | 354 | 287 | 327 | 421 | 412 | 371 |
| 2008 | 292 | 271 | 290 | 284 | 244 | 208 | 147 | 319 | 284 |
| 2009 | 288 | 293 | 266 | 243 | 238 | 233 | 230 | 220 | 251 |
| 2010 | 325 | 320 | 320 | 325 | 443 | 348 | 315 | 280 | 319 |

Productivity (Kg lint per hectare) in Vidarbha

Compiled by Reddy, A. R (2012) CICR, Nagpur



41. Is Bt cotton really controlling bollworms?

Bt-cotton has been highly effective in controlling the bollworms. The Bt-cotton technology gives at least 70-80% protection against bollworms. Initially in the season it gives almost 100% control of the bollworm up to 80-85 days old crop. Later in the season about 10-20% insects can survive on the crop. The Cry1Ac expression was found to be high in leaves and less in flowers and bolls. It was also found to reduce in leaves after 110 days after sowing.

Bt hybrids exhibited higher tolerance to bollworm damage. Bambawale *et al.* (2003) reported a 50% overall reduction in the *H. armigera* larval population in Bollgard-MECH-162 compared to the non-Bt MECH-162. Their data showed that the total per cent damage to fruiting bodies, including squares and flowers, green bolls and shed reproductive parts was 65% lower in Bollgard-MECH-162 compared to non-Bt MECH-162. Further, the locule damage caused by pink bollworm was found to be 58% lesser in Bt-cotton. Udikere *et al.* (2003) also showed that the three Bt-cotton hybrids, Bollgard-MECH-162, Bollgard-MECH-162 and Bollgard-MECH-184 were able to reduce larval populations of *H. armigera* up to 40%, spotted bollworm (*Earias vittella*) up to 30-40% and pink bollworm (*Pectinophora gossypiella*) up to 60-80% in south India.

However, some occurrence of bollworms is reported sometimes from a few fields across the country. This is because of the fact that India is the only country in the world that cultivates hybrids and also that the bolls on F1 hybrid plants possess 25% non-Bt seeds, enables the survival of bollworm larvae that feed mainly on the developing seed. Moreover, boll rind and fruiting parts of Bt-cotton express low levels of Cry toxins, which are sometimes inadequate to kill the larvae.

42. How many sprays are being used now to control bollworms in cotton?

By the mid 1990s Indian cotton farmers were spending >43% of the variable costs of cotton production on insecticides, around 80% of that being for bollworm control and in particular *Helicoverpa* control (ICAC 1998a, b). Insecticide use on cotton was 50% of all insecticide use in the country. Cotton production was being rendered uneconomic in many regions of the country. The reasons for the very rapid increase in the importance of *H. armigera* as a cotton pest are unknown but by the end of the decade it was the major cotton pest. In 1998-99, at least 14.6% of Indian cotton production was lost to insect (mainly bollworm) damage.

Farmers were taking up 15-30 applications of insecticides, mostly tank-mixes. The excessive use of insecticides, especially synthetic pyrethroids, led to further and worse problems of insecticide resistance in *Helicoverpa armigera* and *Spodoptera*



litura, which further necessitated the repeated application of insecticides. The first few reports related to high levels of *H. armigera* resistance to pyrethroids and DDT. Later, several laboratories reported high levels of pyrethroid resistance in several cotton and pulse growing regions of the country. Subsequent studies (Kranthi et al., 2002) showed that resistance to pyrethroids was ubiquitous and resistance in *H. armigera* to conventional insecticides such as methomyl, endosulfan and quinalphos was increasing in India. Due to unsatisfactory insect control on account of insecticide resistance, farmers were forced to spray repeatedly, most often with mixtures. By 1992, *H. armigera* resistance to insecticides had emerged as a great challenge to cotton pest management in Asia and Australia. Similar problems were being experienced in the Americas with other heliothine species. Subsequently, a number of Integrated Pest Management (IPM) programmes were initiated across the world in cotton growing countries, but these were only partly successful in keeping bollworms under check.

After the introduction of Bt-cotton in 2002, insecticide use for bollworm control declined rapidly and currently very few insecticide sprays are used for bollworm control in Bt-cotton.

43. Have there been changes in insecticide usage for bollworm and sucking pest control?

Clearly, insecticide usage for bollworm control decreased after 2004 and usage for sucking pest control increased after 2006.

| | Value | of insectio | ides in Rs | crores | Quantity of insecticides in Metric tonnes | | | | | |
|------|---------------------------------------|--|------------------------------------|-----------------------|---|-----------|-------------|------------------------------------|-----------------------|--|
| | Insecticides used for Bollworms | Insecticides used for Sucking pests | Total insecticides on cotton | Total Insecticides | Sucking pests | Bollworms | Other pests | Total insecticides on cotton | Total insecticides | |
| 1995 | 500.5 | 200.3 | 700.8 | 1288 | 2965 | 5748 | 487 | 9200 | 26923 | |
| 1996 | 524 | 256 | 780 | 1517 | 3643 | 5920 | 492 | 10054 | 29752 | |
| 1997 | 575.5 | 238 | 813 | 1653 | 3621 | 6973 | 361 | 10955 | 27471 | |
| 1998 | 606.3 | 247 | 854 | 1907 | 3857 | 7930 | 418 | 12205 | 31268 | |
| 1999 | 569.3 | 310 | 879 | 2128 | 4487 | 7522 | 320 | 12329 | 33398 | |
| 2000 | 547 | 292 | 839 | 2052 | 3716 | 6647 | 625 | 10988 | 30120 | |
| 2001 | 747.6 | 304 | 1052 | 2268 | 3312 | 9410 | 454 | 13176 | 34910 | |
| 2002 | 415.6 | 181 | 597 | 1683 | 2110 | 4470 | 283 | 6863 | 25962 | |
| 2003 | 680.5 | 245 | 925 | 2146 | 2909 | 6599 | 537 | 10045 | 32571 | |
| 2004 | 718.1 | 314 | 1032 | 2455 | 2735 | 6454 | 178 | 9367 | 35432 | |
| 2005 | 385.7 | 263 | 649 | 2086 | 2688 | 2923 | 302 | 5914 | 32750 | |
| 2006 | 307.4 | 272 | 579 | 2223 | 2374 | 1874 | 375 | 4623 | 31363 | |
| 2007 | 287.8 | 445 | 733 | 2880 | 3805 | 1201 | 536 | 5543 | 35807 | |
| 2008 | 236.7 | 554 | 791 | 3282 | 3877 | 652 | 528 | 5057 | 26624 | |
| 2009 | 140.1 | 694 | 834 | 3909 | 5816 | 500 | 410 | 6726 | 35404 | |
| 2010 | 122.8 | 758 | 880 | 4283 | 7270 | 249 | 366 | 7885 | 36761 | |
| 2011 | 96.3 | 894 | 991 | 4103 | 6372 | 222 | 234 | 6828 | 34469 | |

Compiled by Kranthi & Reddy (2012), CICR

44. Is the pesticide use increasing again now and why?

Pesticide usage is increasing again on cotton in India over the past 5 years. Insecticide usage on sucking pests has increased from 2374 M tonnes in 2006 to 7270 M tonnes in 2010 and 6372 M tonnes in 2011. The value of insecticides used on cotton for sucking pest control was Rs 272 crores in 2006, which increased to Rs 894 crores in 2011. The increase in insecticide use could be due to the following reasons:

- a) Approval of 1128 Bt-hybrids across the country by 2012. Many of these hybrids are susceptible to sucking pests.
- b) Increase in the area under sucking pest-susceptible hybrids
- c) Increase in resistance levels of jassids to Imidacloprid and many other recommended insecticides

Since 2002, every Bt-cotton seed has been treated with the highly effective insecticide 'imidacloprid'. Farmers have also been spraying this insecticide on the crop to control jassids. Jassids have developed resistance to 'imidacloprid' and therefore crop can be damaged and yields are likely to decline due to sucking pests.



45. Have bollworm populations declined because of Bt cotton?

Interestingly, *H. armigera* infestation reduced significantly in cotton ecosystems from 2000, to the point of effective non-existence in some parts of India. It is not clear whether it was the introduction of Bt-cotton or the change in insecticide use pattern in Asia, notably the decrease in pyrethroids, coupled with increase in the new

chemistries which impose fitness problems in residual surviving populations, which caused the change, but *H. armigera* populations rarely exceeded economic threshold levels in Asia, particularly in majority of the cotton growing regions of India. It is now being increasingly felt that bollworm infestations declined significantly over the past 12 years mainly because of a significant decline in the use of the insecticide "synthetic-pyrethroid" coupled with enhanced usage of some potent bollworm-controlling insecticides such as Spinosad, Emamectin and Indoxacarb, which were introduced during 2000-2001. Bt cotton has also played a part in the decline of bollworm populations.

46. Do the bollworms find new host crops because of Bt cotton?

The cotton bollworm *Helicoverpa armigera* is known as a recorded feeder on 181 host plants. Amongst these, pigeonpea, chickpea, tomato, maize, cotton, sunflower and several vegetables are the common crop hosts. Thus far there is no record of any new host of the cotton bollworm after the introduction of Bt cotton in India. Further there is no scientific evidence to show that there has been any increase in the infestation levels of the cotton bollworm on any of the common host crops as a consequence of the introduction of Bt cotton.

47. Have the bollworm populations increased in non-Bt crops?

Bt-cotton does not repel bollworms for them to seek alternate host plants. The Cry1Ac toxin in Bt cotton kills the bollworms and has been effectively controlling the bollworm populations to an extent of more than 90%. Consequently, the bollworm infestation and population levels have significantly decreased on cotton and also on many other host crops. Cotton is the first target crop for the bollworm in Kharif. The bollworm is known to move on to other crops such as pigeonpea and sunflower after completing 2-3 cycles on cotton and subsequently on to chickpea and winter vegetables. The carry-over population from cotton has significantly declined because of effective control by Bt-cotton and thus it is not right to presume that bollworms have found new hosts after the introduction of Bt cotton.

48. Do bollworms still occur on Bt cotton?

Bollworms are often found as younger stages of larvae on Bt-cotton plants. Some plant parts such as the boll rind, square bracts, buds and flowers which express low levels of Cry1Ac, may sustain a small proportion of larvae that feed on them. *In-vivo* and *semi in-vivo* bioassays were conducted on intact plants and isolated plant parts. The assays indicated that a small proportion of larvae survive under field conditions and majority of these grew well on flowers and boll rind. Survival of 5-10% larvae on Bt-cotton plant parts in *semi-invivo* bioassays is not uncommon. An overall analysis revealed that the Bt-cotton technology had a capability of reducing insect pest infestations by 60-90% under field conditions. The efficacy to a large extent was dependent on the host into which Cry1Ac was introgressed.

49. How can bollworms still feed on Bt cotton?

The toxin expression is highest in leaves followed by squares, bolls and flowers. The expression levels in leaves decline after 110-120 days after sowing. Therefore, Bt-cotton controls bollworms effectively at 90-100% up to 100-110 days after sowing and 70-80% of the bollworm larvae thereafter. The cotton bollworm *Helicoverpa armigera* lays majority (70-80%) of its eggs on leaves of the upper canopy and neonate larvae scrape and feed on the surface of the leaf soon after hatching and get killed. However rest of the eggs laid directly on squares, flowers and bolls can survive, depending on the levels of toxin expression in these parts. Hence, at times of high pest pressure, insecticide sprays may become necessary to protect the Bt-cotton crop. The bolls on F-1 plants contain seeds which segregate in 3:1 ratio of Bt:non-Bt. Therefore bollworm larvae may survive on the 25% non-Bt seeds in green bolls if they manage to bore into green bolls. The pink bollworm survival in Bt-cotton is mainly due to the presence of such segregating Bt-cotton seeds in the green bolls of the Bt-cotton F-1 hybrids

50. Is the toxin expression variable in hybrids, plant parts and over the season?

Cry1Ac expression ranged at 0.01 to19 µg/g in various parts of the plant. The highest expression was in leaves at 75 days after sowing (DAS). A decline in expression of toxin levels was observed in all the eight hybrids. The earliest decrease was in MECH-162, with toxin levels falling off to 1-2 µg/g by 85 DAS. Expression in some hybrids such as RCH-144 and MECH-184 declined only after the 120th day after sowing. The expression levels were highly variable in different plant parts. Though younger leaves expressed highest levels of the toxin, there was a lot of variability in expression. The boll rind, buds and flowers had low expression at 0.01 to 2 μ g/g. On an average the Cry1Ac expression in the eight Bt-cotton hybrids was found to be adequate for bollworm protection at least until the first 100-120 days after sowing. However, some plant parts such as the boll rind, square bracts, buds and flowers which express low levels of Cry1Ac, may sustain a small proportion of larvae that feed on them. In-vivo and semi in-vivo bioassays were conducted on intact plants and isolated plant parts. The assays indicated that a small proportion of larvae survive under field conditions and majority of these grew well on flowers and boll rind. Survival of 5-10% larvae on Bt-cotton plant parts in semi-invivo bioassays is not uncommon. Though 2-3 fold differences in Cry1Ac levels were common between the hybrids during early phase of the crop growth, variability up to 7-fold was also observed at times. The current study showed that increasing levels of H. armigera survival were correlated with the toxin levels decreasing below 1.8 µg/g of the plant parts.

Further, it was found that soil nutrient deficiencies, especially of nitrogen lowered the levels of toxin expression. Therefore proper nutrient management of Bt-hybrids also forms an important component of effective bollworm management.



51. What is refugia?

Refugia is a method in which the non-Bt version of the crop is planted in the vicinity of the Bt-crop so as to ensure the survival and maintenance of susceptible insect populations on the non-Bt crop. The strategy is based on the fact that if a small defined area of non-trangenic plants are cultivated in close vicinity of the toxin expressing transgenic plants, they serve as hosts of the target Bt-susceptible insect pests to multiply. These would then serve as reservoirs of the susceptible alleles and when mated with the rare resistant survivors from transgenic plants would result in heterozygous progeny which would express susceptibility, especially if the resistant alleles are recessive in nature. The probability of the susceptible alleles mating with the resistant insects from Bt plants would be high because of the large population of susceptible insects from the non-Bt refuge. Hence having a refuge in close proximity helps in the effectiveness of the refuge.

In India the Genetic Engineering Approval Committee (GEAC) has recommended refuge of non-Bt (5 border rows) with Bt-cotton per acre or an area of 20% Bt cotton that can be subjected to insecticide sprays. Recently, in 2009, pigeonpea has also been approved as refugia to be cultivated as border rows around Bt cotton.

52. Are farmers following the refugia guidelines?

The recommended strategy of ensuring a 5-row non-Bt crop all around an acre of Btcotton crop, did not become popular with farmers. The general feeling was that cultivating non-Bt cotton in 20.0% of the area would make the crop vulnerable to bollworms, thus warranting pesticide usage and possible lowered productivity. The farmer was vexed with the bollworms and was not willing to allow his crop to be exposed to the possibility of bollworm damage and crop losses. In general, the refugia strategy was not followed in the country. Moreover, in small farms of 1 to 2 acres, use of insecticide on the small refuge-patch of 0.2 to 0.4 acres is economically unviable.

53. Will bollworms develop resistance to Bt-cotton?

After the introduction of Bt transgenic cotton in 2002 and large scale cultivation at more than 67.0% from 2007 it is reasonably certain that bollworms, especially the cotton bollworm, *Helicoverpa armigera*, will respond to the intense selection pressure through a decline in its susceptibility to *cry1Ac*, the gene used frequently against it. However, the introduction of Bollgard II in 2006 has helped in delaying bollworm resistance development to Cry1Ac. Bollgard II contains two genes, Cry1Ac + Cry2Ab, which have unrelated mode of action and mechanism of resistance. Deployment of

two unrelated genes together helps in delaying resistance. However, it is important to develop strategies to retard the rate of resistance development.

54. What are the various resistance management strategies being adopted across the globe?

There have been significant changes in the IRM strategies world over. Since, Bollgard II accounted for more than 80% of the annual plantings in Australia over the past few years after 2006, the regulatory authorities of Australia have stipulated the following refugia conditions for Bollgard II. A grower with 100 ha of Bollgard II has four refuge options.

- 1) 10 ha unsprayed conventional cotton,
- 2) 5% irrigated unsprayed pigeon pea,
- 3) 15% irrigated unsprayed sorghum,
- 4) 20% irrigated unsprayed maize.

The refuge field has to be within two kilometers of the Bollgard II crop. In the US, the EPA has approved a natural refuge for Bollgard II for Alabama, Arkansas, Florida, Georgia, Kansas, Kentucky, Lousiana, Missouri, Mississippi, North Carolina, Oklahoma, South Carolina, Tennesse, parts of Texas and Virginia. Thus in effect the natural refuge is operational in all the states east of Texas and in most of the counties of Texas. Refuge at 5% is mandatory in states/ counties which are endemic for pink bollworm. The natural refuge option gives growers a choice to use alternate host crops instead of conventional cotton for refuge purposes.

55. How is bollworm resistance monitored?

The changes in variability of the cotton bollworm, *Helicoverpa armigera* susceptibility levels to the *Bacillus thuringiensis* Bt toxin (Cry1Ac) were monitored through log dose probit assays conducted on populations collected from major cotton growing districts of India during the 10 year period from 2002-2012.

Every year, bollworm larvae or eggs are collected from different cotton growing districts across the country. While eggs are collected from any crop including Bt-cotton, larvae are collected only from non-Bt crops, so as to ensure that they are not the result of selection pressure. Bollworm larvae were collected from non-Bt cotton crop, pigeonpea and chickpea. Wherever non-Bt cotton crop was unavailable, eggs were collected from Bt crop. The larvae were reared on semi-synthetic diet until pupation and subsequent moth emergence. Bioassays were conducted on the first generation one-day old larvae using Cry1Ac toxin incorporated into semi-synthetic diet. The Cry1Ac toxin used in the bioassays was identical to the protein expressed in Bt cotton.

Twenty to twenty four larvae were used per concentration in three replications on 5 graded concentrations ranging from 0.001 to 5.0 μ g Cry1Ac/ml of diet. All the larvae were kept individually to prevent cannibalism. The toxin treated diet was changed twice during the bioassays. Growth inhibitory and mortality observations were



recorded for 6 days. Weights of surviving larvae were recorded on the final day of observation. The assays were performed in the laboratory at conditions of 27 $\pm 1^0 C$ and 70% relative humidity. Median Lethal Concentrations (LC₅₀) and Median growth inhibitory concentrations (EC₅₀) were derived from log dose probit calculations.

56. Have bollworms developed resistance to Bt-cotton so far?

Studies carried out by CICR (Kranthi et. al., unpublished) showed that there was a decline in the proportion of susceptible populations. Summary of resistance monitoring to Cry1Ac toxin of Bt cotton on cotton bollworm populations collected from various locations in India. The **LC**₅₀ and **IC**₅₀ are expressed in μ g Cry1Ac/ml of diet. RR is resistance ratio in comparison with the reference susceptible strains.

| Sites | Highest IC ₅₀ | Resistance Ratio | Highest LC ₅₀ | Resistance Ratio |
|-------|--|---|--|--|
| 10 | 0.034 | 2 | 0.67 | 7 |
| 45 | 0.043 | 2 | 0.54 | 5 |
| 20 | 0.023 | 1 | 0.38 | 4 |
| 21 | 0.104 | 5 | 0.74 | 7 |
| 39 | 0.166 | 9 | 0.72 | 7 |
| 27 | 0.195 | 10 | 0.79 | 8 |
| 49 | 0.201 | 11 | 1.15 | 12 |
| 26 | 0.58 | 31 | 3.12 | 31 |
| 31 | 0.59 | 31 | 3.14 | 31 |
| 27 | 0.24 | 13 | 3.26 | 33 |
| 17 | 0.36 | 19 | 5.10 | 51 |
| | Sites 10 45 20 21 39 27 49 26 31 27 17 | HighestSitesIC50100.034450.043200.023210.104390.166270.195490.201260.58310.59270.24170.36 | Highest IC50Resistance Ratio100.0342450.0432200.0231210.1045390.1669270.19510490.20111260.5831310.5931270.2413170.3619 | Highest IC50Resistance RatioHighest LC50100.03420.67450.04320.54200.02310.38210.10450.74390.16690.72270.195100.79490.2011111.15260.58313.12310.59313.14270.24133.26170.36195.10 |

Data (Kranthi, unpublished)

The LC₅₀ (median lethal concentration) values ranged from 0.02 to 0.54 µg Cry1Ac/ml of diet in 2002 and 0.246 to 5.10 μ g Cry1Ac/ml of diet in 2011-12. The IC₅₀ values (median growth inhibitory concentration) ranged from 0.003- 0.034 µg Cry1Ac/ml of diet in 2002 and 0.036 to 0.363 µg Cry 1Ac/ml of diet in 2011-12. Susceptible populations were maintained in the laboratory and used in bioassays as reference during all the years. Resistance ratios (RR) were derived in comparison with the reference susceptible data. To ensure a sustained method of comparison across the years, reference IC₅₀ values of 0.019 µg Cry1Ac/ml and LC₅₀ value of 0.1 µg Cry1Ac/ml of diet of the standard susceptible strain were used to derive the RR (Resistance Ratio) values across the years. While more than 90% of the populations showed typical susceptible response of low LC_{50} and $1C_{50}$ values, resistance ratios of 31-fold were recorded in one or two locations during 2008-09 to 2010-11 and 51-fold in one location during 2011-12. However, Bt cotton was found to be effective in controlling bollworms in the districts wherein the resistance ratios were 51-fold. Thus it appears that the data did not indicate levels of resistance in the populations that may be adequate for significant survival of the populations under field conditions in any of the populations tested. However, the data indicated that there was a clear decrease in the proportion of susceptible populations. For example, the proportions of populations exhibiting LC₅₀ values of less than 0.1 μ g Cry1Ac/ml of diet were 22-38%

between 2002 and 2004, which decreased to 5-12% during 2005 to 2008. Similarly, the proportions of populations exhibiting LC_{50} values of less than 0.01 µg Cry1Ac/ml of diet, were 22-50% between 2002 and 2003, which decreased to 0-3% during 2004 to 2008. In general majority (more than 90%) of the populations exhibited a normal susceptible response in bioassays with resistance ratios less than 10-fold.

It can be surmised from the 10 year monitoring data that, thus far that majority of the bollworm populations collected and tested from various locations across the country are susceptible to Bt-cotton and the few populations that showed up to 51-fold decrease in susceptibility were still being controlled by Bt-cotton under field conditions.

57. Has the pink bollworm developed resistance to Bt-cotton in India?

Monsanto issued a press release in the first week of March 2010 stating that 'during field monitoring of the 2009 cotton crop in the state of Gujarat in western India, Monsanto and Mahyco scientists detected unusual survival of pink bollworm to first-generation single-protein Bollgard cotton. Testing was conducted to assess for resistance to Cry1Ac, the Bt protein in Bollgard cotton, and pink bollworm resistance to Cry1Ac was confirmed in four districts in Gujarat – Amreli, Bhavnagar, Junagarh and Rajkot. Gujarat is one of nine states in India where cotton is grown. To date, no insect resistance to Cry1Ac has been confirmed outside the four districts in Gujarat.'

58. What kind of approaches can be adopted to delay resistance?

Some of the universally proposed strategies include; use of multiple toxins, rotation of toxin genes, crop rotation, seed mixes, gene pyramiding, high or ultra high dosages, and spatial and temporal refugia. A combination of more than one or all of these tactics together may also prove beneficial in attempts to prevent or diminish the selection of rare individuals carrying resistance genes. Amongst the several strategies recommended worldwide, refugia has been one of the most commonly deployed resistance management strategies.

In light of the facts that the Cry1Ac expressed in the current Bt cotton events does not represent 'high dose' against *H. armigera* and also that the allele conferring bollworm resistance to Cry1Ac, is not extremely rare and is inherited in a semi-dominant manner, it is important to develop resistance management strategies appropriate for Indian conditions. More importantly, the strategies should be acceptable to the Indian farmer and should be compatible with the existing cropping systems and management practices. Resistance management approaches generally rely on (1) Conserving susceptibility by minimizing toxin exposure or (2) getting rid of resistant RS and RR genotypes by using either high dose of the same toxin or by using other unrelated toxins.

IRM strategies for India should focus more on the deployment of gene stacks such as the one (Cry1Ac+Cry2Ab) present in Bollgard II, which has toxin combinations with different modes of action and different mechanisms of resistance and therefore very

small likelihood of cross-resistance. Other strategies such as non-Bt cotton or pigeonpea as refugia and control of residual larvae on Bt cotton using bio-pesticides are useful options to delay the onset of resistance and ensure that the benefits of the technology are harnessed for the longest possible time.

59. Do we have 'India-specific' IRM strategies to delay resistance?

IRM strategies suggested for India

- a) Early duration pigeonpea variety as border rows
- b) Cotton varieties or hybrids of matching fiber quality and phenotype can be used as 5-20% refugia
- c) Staggered planting of 1% area under intermittent rows of okra that can act as refugia for all the three bollworms.
- d) One bio-insecticide or insecticide spray at 100-150 DAS for bollworm control in Bt cotton to minimize survival of Bt resistant larvae
- e) Gene stacks with extremely small likelihood of cross resistance. New GM products must be based on two or more independently acting genes in a stack
- f) Hand-picking of surviving larvae from Bt-cotton fields during September in North, October in Central & November in South India.
- g) Destroy residual pupae by deep ploughing in Bt-cotton fields immediately after final harvest
- h) Timely crop termination & destroy stalks
- i) Phase out single gene products as soon as possible
- j) To enhance practicability, 5% Refuge in bag (same non-Bt hybrid) may be recommended + 5% pigeon-pea plants as a patch.



Pigeonpea Refugia

Chapter - 8 NEW GM COTTON TECHNOLOGIES

60. Are there new Bt genes for new versions of Bt-cotton?

Monsanto's Bt-cotton technology Bollgard-II contains Cry1Ac + Cry2Ab. Dow agro Sciences are conducting trials with Wide-strike (Cry1Ac + Cry1F + pat); Bayer, India have initiated trials with twin-link (Cry1Ab + Cry2Ae + pat) and JK seeds have started trials on Cry1Ac+Cry1EC. The *vip3A* gene is yet another toxin that is likely to be pyramided with the existing toxins.

61. Are we going to continue with addition of new genes for insect control?

Continuous addition of new genes may not be technologically feasible and commercially viable, either for the companies or for the farmer. Ideally, it would be most appropriate to create blends of conventional resistant sources with the GM traits so that the technology becomes more durable, sustainable and cost-effective.

Monsanto and Dow, together have stacked eight genes into GM-Maize (corn) called SmartStax. Six of these are for insect control, Cry1A.105, Cry2Ab2 and Cry1F for borers; Cry3Bb1, Cry34Ab1 and Cry 35Ab1 for root worms; *pat* for glufosinate resistance and CP4 EPSPS for glyphosate resistance. The technology of gene pyramiding is commendable, but it will have to sustain economic and ecological tests in the near immediate future.

62. What is herbicide resistant cotton?

The new herbicide resistant cotton technology being introduced by Monsanto in India is called 'Roundup Ready-Flex'. The advantage with the technology is that farmers can spray the broad spectrum herbicide glyphosate (Round-up) on cotton crop directly, which results in weeds getting killed but without any adverse effect on the cotton crop. Thus the GM technology makes the herbicide selective to weeds without affecting the crop itself. Bayer Crop Science is initiating field trials of glyphosate and glufosinate resistant GM cotton in India. Dow agro sciences are conducting field trials of glufosinate resistant GM cotton in India.

63. Can herbicide resistant cotton be grown with inter-crops?

The RRFlex technology or any other herbicide resistant GM technologies, do not support cotton intercropping with the commonly used inter crops such as pigeonpea, soybean, maize, jowar etc., which were cultivated as part of risk aversion or sustenance. Moreover, reduction in area of inter-crops can hasten development of bollworm resistance to Bt-cotton. Alternative plans need to be designed



64. What have been the benefits of Bt-cotton so far in India?

Direct benefits from Bt-cotton in India

The main purpose of Bt-cotton is to control bollworms.

Bollworm control: Bt cotton effectively controlled bollworms, especially the American Bollworm, *Helicoverpa armigera,* thus preventing yield losses from an estimated damage of 30.0 to 60.0% each year in India thus far from 2002 to 2011.

<u>Increased yields:</u> Yields are estimated to have increased at least by 30.0% due to effective protection from bollworm damage.

<u>Reduction in pesticide use for bollworm control:</u> The biggest gain from the technology was in the form of reduced insecticide usage from 46% in 2001 to less than 26% after 2006 and 21% during the last two years 2009 and 2011. Prior to the introduction of Bt cotton, about 9400 M tonnes of insecticides were used for bollworm control in India. In 2011, only 222 M tonnes were used for bollworm control.

<u>Reduction in bollworm infestations:</u> The intensity of bollworms reduced significantly on cotton and also on other host crops.

Elimination of bollworm threats: Farmers are no longer scared of impending bollworm infestations and the subsequent stress of using insecticide cocktails.

<u>Enhanced seed-cotton quality:</u> The quality of seed-cotton from Bt-cotton fields was found to be better than non-Bt cotton because of negligible loculi damage and fiber damage.

Earliness and determinate habit: Introduction of Bt gene into the hybrids has added the advantage of protection of early fruiting parts, thus resulting in earliness and determinate habit. The earliness ranged from 15 to 20 days in many hybrids in many parts of the country. There have been several added benefits to this. In North India, farmers were able to take up wheat cultivation immediately after early harvest of cotton. The number of picking reduced and the yield per each of the few pickings, increased. Farmers were able to get remunerative returns because of higher prices generally prevalent early in the market during the initial cotton arrivals.

65. Has there been any environmental benefit with Bt-cotton?

The greatest environmental benefit with Bt-cotton is the reduction in insecticide usage. The average insecticide usage for bollworm control over 10 years from 1995 to 2004 was 6767 M tonnes, which reduced to an average of 1089 M tonnes over seven years from 2005 to 2011. However the average usage of insecticide for sucking pest control was 3335 M tonnes during 1995 to 2004, which increased to an

average of 4600 M tonnes during 2005 to 2011. Though this is not directly related to Bt cotton technology, the pesticide scenario could have been very different, with pesticides reduced to low levels if care had been taken to ensure that sucking pest resistant hybrids were approved for commercial cultivation.

66. How can we improve the environmental benefits?

Sap-sucking pest resistant Bt-cotton varieties can certainly contribute to immense reduction in the overall usage of insecticides for sucking pests and bollworms, thereby enhancing naturally occurring biological control and environmental benefits.

67. What have been the benefits of Bt-cotton, so far, abroad?

Bt-cotton has been successful so far in protecting cotton crop against bollworm damage. Effective protection resulted in enhanced yields and reduced need for pesticide use for bollworm control.

In India the cultivation of Bt cotton has reduced pesticide application to 41% and increased the effective yield to 37% there by resulted in the increase of profit to approximately Rs. 6000/- per hectare. Kathage and Qaim (2012) collected unique panel data between 2002 and 2008 in India, with nonrandom selection bias in technology adoption and showed that Bt has caused a 24% increase in cotton yield per acre through reduced pest damage and a 50% gain in cotton profit among smallholders. They found that these benefits are stable; there are even indications that they have increased over time. They further showed that Bt cotton adoption has raised consumption expenditures, a common measure of household living standard, by 18% during the 2006–2008 period. They concluded that Bt cotton has created large and sustainable benefits, which contribute to positive economic and social development in India

China obtained the highest profit to around Rs. 21,000/- per hectare reducing the pesticide application to 65%.

In Mexico the reduction of pesticide was 77% resulting in the profit of around Rs. 13,000/- per hectare.

Yield increase of the cotton in top five major cotton growing countries before and after introduction of GM cotton revealed that there is significant yield gain. In India, 82% increase in yield has been noticed followed by 49% in China, 42% in USA, 29% in Australia and 7% in Pakistan.

68. Has the quality of cotton improved with Bt-cotton?

The quality of seed-cotton from Bt-cotton fields was found to be better than non-Bt cotton because of negligible loculi damage and fiber damage. The quality improved because of reduced bollworm damage. Trash content was reduced and bollworm affected 'bad-kapas' was extremely low. Bt cotton hybrids showed higher retention of

first formed bolls due to low fruiting point and boll damage thus exhibiting more balanced plant growth.

Prior to 2002 long staple cotton production was only 38% of the total cotton, but the proportion increased to 77% by 2007. The quality of Indian cotton which was hitherto considered as inferior, is now acceptable internationally as export quality with improvement in quality after the introduction of Bt cotton. Bt cotton did not have any adverse effects on fiber quality. However, the textile industry pointed out that micronaire (fineness) value declined in the later pickings. Due to early retention of bolls in Bt cotton hybrids, the boll bursting commenced nearly 15-20 days in advance and required lesser number of pickings to complete the harvest.

69. Did our imports decline and exports increase with Bt-cotton introduction?

India has been producing at least 1.0 M metric tonnes in excess of domestic consumption over the past few years. Domestic consumption also increased from 2.87 M tonnes in 2002, to 4.52 M tonnes in 2010.

India became a leading global exporter of raw cotton with exports averaging at 53 lakh bales over nine years from 2003-2011 compared to an average of 1.18 lakh bales during the years 1997 to 2002 prior to the introduction of Bt cotton. Imports declined from an average of 16.50 lakh bales over 6 years between 1997 to 2002, to an average of 6.9 lakh bales over 9 years from 2003 to 2011. India exported a record 129 lakh bales in 2012.



Indian Markets with surplus cotton

Chapter - 10 CRITICISMS AND CONCERNS

70. What has been negative fallout of the cultivation of Bt cotton on the farmers field?

The public sector varieties and hybrids, which are intensively tested for agroecological suitability for specific regions, prior to approval, are no longer being used by farmers, because they do not have Bt gene in them. It is interesting to note that only 40 intra-*hirsutum* hybrids were released by the public sector over 40 years from 1970 to 2010, after rigorous field testing. But, more than 1000 Bt-cotton hybrids were approved by the GEAC (Genetic Engineering Appraisal Committee) in five years from 2007 to 2012.

The area under Desi cotton was 97% in 1947; 42% in 1990; 28% in 2000 and is estimated to be less than 3% in 2011. Desi cotton species *G. arboreum* are native to India and are highly resistant to drought, insect pests and diseases. Desi cotton is grown only in India and several high yielding short staple coarse fiber varieties are best suited for surgical cotton. India could have easily created a huge exclusive global market for 'surgical cotton' and 'organic-surgical-cotton' using the native Desi varieties. The fields in which Desi cotton varieties were cultivated were either saline or marginal and unsuitable for hybrid cotton. The cost of cultivation of Desi cotton is low since the requirement for pesticides and fertilizers is low.

A total of 1128 Bt-cotton hybrids have been approved for commercial cultivation by 2012, thus creating confusion among farmers.

With the availability of innumerable hybrids, seed testing agencies are finding it difficult to monitor the quality of seed. Thus, seed quality has become a major issue. It has also given rise to sale of spurious seed.

With seed shortage of popular hybrids, farmers are persuaded to try new hybrids every year.

With the entry of Bt cotton, the practice of use of 'farm saved seeds' declined.

High seed cost is an issue, especially in cases of poor germination arising out of low soil moisture and erratic onset of monsoon.

Most of the Bt cotton hybrids available are susceptible to the sucking pests, thus necessitating increase in pesticide usage.

Because of the increased cost of labor in hybrid seed production, it was alleged that some seed producers were employing child labor for hybrid seed production to reduce the costs. However, stringent care is now being taken by the Government and also by the seed producers to ensure that child labor is not used at any stage for cotton cropping or seed production.

71. What are the major criticisms of Bt-cotton by NGOs and concerns expressed by farmers?

Right from the inception of the technology there has been a sustained opposition from some of the NGO groups. The initial opposition was very speculative and confusing without any reasonable assessment of the technological strengths of Bt-cotton. Most of the criticism was also based on plain ignorance. The Karnataka Ryta Sangha conducted public demonstrations against Bt-cotton and uprooted a few Bt-cotton experimental plots in 1998 and 1999 with misleading accusations of the possible presence of the 'terminator' genes in Bt-cotton. Later several NGOs started highlighting crop failures as failure of Bt-cotton technology. Clearly crop failures resulting from either abiotic or biotic stress, were being attributed to Bt-technology, ignoring the fact that Bt-cotton was developed specifically to offer protection against bollworms, not against any other adverse factors.



parawilt affected plant

Leaf reddening

However, issues related to the occurrence of bollworms on Bt-cotton and the increasing levels of *Spodoptera litura* on Bt cotton are related to the Bt technology and must be addressed.

There have been several problems pointed out by NGOs and farmers. Some of them are being listed below:

- a) Increase of insecticide usage on Bt-cotton
- b) Death of sheep, goats and cattle after feeding on Bt-cotton
- c) Bt-cotton was reported to be more susceptible to jassid and thrip infestation

- Emergence of mealybugs, mirid bugs and gall midges as a problem on Btcotton
- e) Leaf streak virus in Bt-cotton was reported in some parts of Andhra Pradesh
- f) Increase in the CLCuD (Cotton leaf curl virus disease) in North India
- g) Problems of leaf reddening and sudden wilt in Bt-cotton fields
- h) Low yields in Bt-cotton in rainfed regions of the country
- i) Re-emergence of *Spodoptera* spp. populations on Bt-cotton
- j) Some Bt-cotton plants harbored the cotton bollworm, Helicoverpa armigera
- k) Pink bollworm infestation was reported to occur on Bt-cotton

72. Is parawilt associated with Bt-cotton?

Parawilt was a problem that was known to occur in cotton in the mid 1970s. A few hybrids were found to be susceptible to the sudden-wilt symptoms. A number of explanations were propounded at that time. The problem soon disappeared over time.

However with the introduction of a few new hybrids, the problem resurfaced again and farmers were incorrectly associating Bt-cotton with parawilt. Parawilt was found to occur due to asphyxiation and can be more in Bt cotton plants because of higher boll retention. Farmers need to be educated that water, nutrient and soil management are extremely critical to get the best performance from Bt-cotton. Poor soils and rainfed conditions are not ideal for the performance of Bt-cotton. It needs optimum water and nutrients at a time when it holds maximum fruiting bodies including green bolls. Bt cotton does not withstand moisture and nutrient stress, especially because the boll retention capacity is much higher as compared to non-Bt varieties. Problems of wilt are generally reported commonly with Bt-cotton. It is true that wilt can be more in Bt-cotton as compared to non-Bt cotton. This is because of the high boll load in Btcotton crop.

- a) Bt cotton suffers from extra moisture and nutrient stress, especially because the boll retention capacity is much higher as compared to non-Bt varieties.
- b) Drought followed by rainfall lead to a 'hiccup' like phenomenon, in the morethirsty Bt-cotton crop, which creates asphyxiation like condition in roots, thus leading to 'parawilt'. Parawilt is more common in shallow soils and rainfed conditions, wherein the performance of Bt-cotton falls below expectations.
- c) Parawilt affected plants also suffer secondary infection from diseases. But this had nothing to do with Cry1Ac or Bt-cotton.
- Studies showed that fungi, bacteria and nematodes were not involved in sudden wilt, but flagellate protozoans in the phloem of wilted plants were observed (Mayee, 1997)
- e) Farmers need to be educated that water, nutrient and soil management are extremely critical to get the best performance from Bt-cotton. Poor soils and rainfed conditions are not ideal for the performance of Bt-cotton. It needs optimum water and nutrients at a time when it holds maximum fruiting bodies including green bolls.
- f) The soil condition must be improved by adding 25-30 tonnes of FYM before sowing. If conditions of parawilt are in the initial stages, drenching the soil

around the infected plants with 1% Bavistin will ward off secondary infection and can help plants in recovery.

73. What are the perceived risks with Bt-cotton?

Bt-cotton actually protects farmers against bollworm risks. Bt-cotton also protects the farmer, consumers, ecology and environment from pesticide risk. Bio-safety tests with Bt-cotton were reasonably robust across the world and have minimized any possible risk to farmers and consumers.

74. Are new pests and diseases appearing on cotton and why?

Bt-cotton is toxic to bollworms and does not control any of the sucking pests of cotton. The Bt-cotton currently released in India is only moderately toxic to the leaf eating caterpillar *Spodoptera*. Over the past seven years, coincidentally after the introduction of Bt-cotton, cotton cultivators in India have been facing new problems with insect pest management in many parts of the country, mostly presumed to be a consequence of low insecticide usage. New sucking pests have emerged as major pests causing significant economic losses.

Though debatable, it is also one of the possibilities that, since the donor parent Coker 312 is known to be highly susceptible to sucking pests such as jassids and thrips, the hybrids may be showing slightly enhanced susceptibility to these pests due to linkage drag, especially if the recurrent parent did not possess inherent resistance to the sucking pests.



Mirid bug on cotton

Mealybugs on cotton boll

It is known that the usage of synthetic pyrethroids for bollworm control had significant negative impact on the incidental populations of *Spodoptera* spp. and several other miscellaneous bugs including the mirid bugs, *Creontiodes biseratence* (Distant), *Ragmus* sp. The reduction of pyrethroids and several conventional insecticides on Bt-cotton is presumed to have led to an enhanced infestation of several non-target

species such as mirid bugs, mealy bugs, thrips and *Spodoptera litura*. Apart from the reports of enhanced disease problems such as grey mildew, leaf spots and rust, new reports of damage by safflower leaf caterpillar in Maharashtra and gall midge damage in Karnataka in cotton, were alarming.

The mealybug was detected as a new species *Phenacoccus solenopsis*, which was not known to be found on cotton in India. Because the problem is new to cotton, it appears to have sent panic signals within the scientific community. There has been a sudden increase in the use of insecticides on cotton, especially those of the extremely hazardous category, over the past two years for mealybug control. Despite insecticide use, the pest was found to spread rapidly all across India causing damage in Punjab, Haryana, Rajasthan, Gujarat and parts of Madhya Pradesh and Maharashtra and is expected to cause more damage if proper precautionary measures are not initiated.

75. Why is it that the cotton leafworm *Spodoptera litura* is able to feed on Bt-cotton?



Spodoptera litura larva

Spodoptera litura moth

The Bt-cotton with Cry1Ac is only moderately toxic to the leaf eating caterpillar *Spodoptera*. Bollgard-II has Cry2Ab which expresses at high levels, and has better control efficacy, compared to the Bt-cotton with only *cry1Ac* gene. However, considering the moderate toxicity of Cry1Ac and Cry2Ab to *Spodoptera litura*, it is likely that the pest may be able to feed and survive on Bollgard II in the next few years. It would not be surprising to see that farmers in some parts of the country would find *Spodoptera* on Bt-cotton.

76. How are these new diseases and insect pests related to Bt-cotton?

The new diseases such as leaf streak virus and new insect pests such as mealybugs, mirid bugs, safflower caterpillars, gall midges etc., are not related to Bt-technology. Other problems such as leaf reddening, para-wilt are also not related to the Bt-technology. These have been occurring because of the susceptible hybrids that were approved as Bt-hybrids for commercial cultivation in the country.

77. How much of Bt toxin gets accumulated in the soil?

The accumulation of Cry toxins in the soil have found to be at negligible levels. Head et al. (2002) demonstrated that the amount of Cry1Ac protein accumulated as a result of continuous use of transgenic Bt cotton, and subsequent incorporation of plant residues into the soil by post-harvest tillage for 3 to 6 consecutive years was extremely low and did not result in detectable biological activity.

78. Have the soils become depleted of nutrients because of Bt-cotton?

Progressive nutrient (Macro and Micro) depletion occurs due to continuous repeated cultivation of high-nutrient consuming hybrids on the same soils, without replenishing nutrients appropriately. The nutrient depletion is not related to the Bt-technology. The source sink relationship gets affected because of repeated hybrid cultivation. Bt-cotton hybrids utilize more nutrients to yield more. Therefore the soils are getting progressively depleted and need more nutrient refurbishment, which is not done properly in many farms. Thus cotton crop shows nutrient deficiency symptoms in many regions, especially in rainfed zones where wilt and leaf reddening problems are getting severe over the years

79. Are the Cry toxins harmful to soil insects?

There are no reports that the Cry1Ac soil-exudates from Bt-cotton plants have any adverse effects on any of the soil insects. Studies conducted in India show that there were hardly any differences in the soil insect profile because of Bt-cotton cultivation.

80. Has Bt-cotton adversely affected soil microbes and soil health?

Reports from India

Microbial community composition and dynamics is an important descriptor for Soil quality alterations. Kapur et al. (2010) assessed the culturable and non-culturable microbial diversities in Bt cotton and non-Bt cotton soils to determine the ecological consequences of application of Bt cotton. Their study indicated that Bt cotton had no effect on the diversity of microbial communities. Diversity of experimental fields was similar during the cropping of both Bt cotton and non-Bt cotton.

Sarkar et al. (2009) concluded from their study that there were no negative effects of Bt-cotton on the soil quality indicators (microbial biomass carbon, microbial biomass nitrogen, microbial biomass phosphorus, total organic carbon, microbial quotient, potential N mineralization, nitrification, nitrate reductase, acid and alkaline phosphatase activities) and therefore cultivation of Bt cotton appears to pose no risk to soil ecosystem functions.

Balachandar et al. (2008) studied the diversity richness of Pink-pigmented facultative methylotrophs (PPFMs) present in the phyllosphere, rhizoplane and internal tissues with Bt and non-Bt cotton. They observed no differences between Bt- and non-Bt cotton.

Based on the three year study (2008 to 2011) conducted at CICR, it was found that growing Bt cotton does not affect the soil biological properties (soil respiration, urease activity, dehydrogenase activity, and microbial biomass carbon). The results obtained with culturable microbial population and microbial diversity index analysis further proved that the microbial activity in soil was not affected by Bt cotton. These results suggest that cultivation of Bt cotton expressing *cry1Ac* gene may not pose any ecological or environmental risk.



Studies conducted abroad:

Rui et al. (2005) reported that the fortification of pure Bt toxin into rhizospheric soil did not result in significant changes in the numbers of culturable functional bacteria, except the nitrogen-fixing bacteria when the concentration of Bt toxin was higher than 500 ng/g. The results indicated that Bt toxin was not the direct factor causing decrease of the numbers of bacteria in the rhizosphere, and other factors may be involved.

Hu et al. (2009) reported that there were no consistent statistically significant differences in the numbers of different groups of functional bacteria between rhizosphere soil of Bt and non-Bt cotton in the same field, and no obvious trends in the numbers of the various group of functional bacteria with the increased duration of Bt cotton cultivation. These studies suggest that multiple-year cultivation of transgenic Bt cotton may not affect the functional bacterial populations in rhizosphere soil.

81. How can we harness the full benefits of Bt-cotton in India?

Bt-cotton technology is only expected to protect the cotton crop from bollworm attacks. Clearly the yield benefits should come from the hybrids or varieties. For best yield benefits, it is important that the hybrids or varieties must be ideally suited for specific agro-eco-zones. For example the best options for rainfed regions would be early-maturing short duration straight varieties, resistant to sucking pests, dwarf statured, zero-monopodial, which are amenable for high-density planting at populations of 100,000 per acre or more. For deep-black soils and irrigated regions, sap-sucking-pest resistant long duration hybrids can be an ideal option.

82. Why do we have Bt-cotton only as hybrids in India?



The technology providers such as Monsanto, JK seeds, Metahelix and Nath seeds and private seed companies in India preferred Bt hybrids in India as a means of "value capture", since farmers cannot reuse the seeds.

In all other countries where the number of farmers is small, the technology providers transferred the genes into the local straight varieties and an agreement form is signed by farmers that they would not reuse the seeds.

In China and Pakistan, where the number of farmers is huge, the agreement form system was not acceptable.

China's public sector research system succeeded in developing its own Bt cotton in straight varieties and went ahead with commercialization.

The argument put forth by the seed sector was that 'hybrid-technology' represents a superior high yielding concept and it is most appropriate to take it forward with Bt-technology to obtain high yields in the country.

83. Which countries other than India cultivate hybrid cotton?

China and Pakistan experimented with Bt-cotton hybrids, but appear to have decided in favor of straight varieties. The area under Bt-hybrids is negligible in these countries now. Thus, India happens to be the only country all across the world to cultivate hybrid cotton and that too, in more than 95% of its cotton area.

84. Do the new GM technologies have potential to increase yields in India?

The new technologies (CP4 EPSPS in Roundup-Ready-Flex, Cry1Ab + Cry2Ae, Cry1Ac + Cry1F and vip3) are expected to be introduced in the next 5-10 years. However these can add to the efficacy of control, but are unlikely to contribute to any additional yield enhancement. No major changes in the yield enhancement or crop protection technologies are expected in the immediate near future. Drought resistance (may be commercially available by 2020). Jassid resistance (through lectin genes may be commercially available by 2020). Leaf curl virus resistant (may be commercially available by 2020).

85. What is the advantage of straight varieties over Bt-hybrids?

The major advantages with Bt-varieties over Bt-hybrids are

- a) The transgenes or cry toxins will be in homozygous condition in varieties, whereas in hybrids these are in hemizygous condition, currently. Studies conducted at CICR showed that homozygous varieties expressed higher levels of toxin as compared to the hemizygous hybrids.
- b) The cry toxins in the Bt-varieties would not segregate in the seeds of the bolls whereas in the bolls of F-1 Bt-hybrids, the seeds would segregate for the Cry toxin at 25.0% non-Bt seeds for a single gene. Thus making the bolls vulnerable to bollworm feeding.
- c) The seeds obtained from straight varieties can be saved and re-used by farmers.
- d) Straight varieties can be sown at high densities of 20-30 times more than the hybrid plant density, to obtain higher yields, especially in rainfed regions and marginal soils. High density of 100,000 plants per acre or more is not possible with hybrids due to prohibitively high seed cost.
- e) Plant population cannot be increased with hybrids. Hybrids are highly input intensive and more susceptible to pests and diseases and thus require more fertilizers and pesticides for optimum production. The cost of hybrid seed is much higher and plant growth is luxuriant and therefore does not permit high density planting.
- f) Specifically developed elite Bt-varieties suitable for high density planting need to be developed on priority.

Chapter - 12 BT AND ORGANIC COTTON

86. Is Bt-cotton compatible with organic cotton?

Currently GM cotton is not accepted by many organic cotton users, especially in Europe. Bt-cotton only expresses a Cry protein and an antibiotic resistance marker protein. Therefore, the technology should have acceptance. However, the explanation for opposition to Bt-cotton as organic crop is either unacceptability of the GM philosophy itself or the assumption that GM crops are cultivated under chemical intensive cultivation.

87. Does Bt-cotton contaminate organic cotton?

If Bt-cotton is cultivated in the immediate vicinity of 10 meters, the border rows may get contaminated. Cotton pollen is heavy and chances of cross pollination by wind are generally extremely low. Pollination by honey bees is possible.

Theoretically, rapid adoption of Bt cotton cultivars into all the cotton growing agroclimatic zones endangers organic cotton movement in their traditional niches as well as spread of organic cotton cultivation to new areas. However, before the introduction of Bt cotton in India in 2002, India was a non-player in the global 'organic cotton' arena, but recently, despite the over whelming presence and steady growth of Bt cotton, India has emerged as a major producer of organic cotton over the past five years (2006-2011).

Currently India contributes more than 70% of the worlds organic cotton produce. India was only a minor organic cotton growing country before 2002 with negligible production of organic cotton. But in 2010, India was the world leader with a contribution of 61% of the organic cotton global production of 175 thousand metric tonnes. About 117,000 farmers are currently engaged in organic cotton production in more than 150,000 hectares in India. The leading states are Madhya Pradesh, Maharashtra and Orissa.

Ironically, this revolution in organic cotton production is occurring in the Bt cotton era wherein 80-85% of the cotton area is under genetically modified (GM) cotton. Thus, India is a classical case of coexistence of GM and organic cotton production systems. Over the past two to three years, India has taken a quantum leap in organic cotton production and has emerged as the global leader in organic cotton production in a dramatic manner, leaving behind Turkey, Syria, China and the USA, who were the leading organic cotton producers until 2007. Dedicated organic cotton growing groups have been cultivating organic cotton by taking complete care of maintaining isolation distance of at least 50 meters from Bt cotton fields, by selecting such villages where this is possible.

88. Are there alternate technologies apart from Bt cotton for pest management?

There are several technologies for cotton pest management that can be used as alternatives to Bt cotton. However, none of them is as efficient as Bt-cotton. The greatest advantage with Bt-cotton is that there is a continuous protection of the crop from bollworms, which are known to cause maximum damage.

All the alternative methods are subject to conditional efficacy and depend largely on weather conditions for application and subsequent effectiveness. The window period becomes small since they rapidly degrade necessitating the need for repeated applications, thus increasing costs. As of now, Bt-technology is the most effective, efficient and environment-friendly options of all available technologies for bollworm management.

Organic cotton can be cultivated profitably only as long as bollworms do not occur in an outbreak form. There are very few eco-friendly technologies to manage pink bollworm and the spotted bollworms.



Chapter - 13 ILLEGAL BRANDS & SPURIOUS SEEDS

89. Can the farmers detect Bt-cotton seeds or plants?

When 'Bt cotton' was first commercialized on 5th April 2002 by Mahyco in India, the initial seed cost was high. 450 gms of the original Bt cotton seed costed Rs 1350 to 1650, About two years prior to the approval of Bt-cotton illegal trade of Bt-cotton seeds was happening in some parts of Gujarat. The illegal versions costed Rs 400 to 600. Lured by the low cost of fake Bt seeds, farmers would easily be trapped into buying them. But they would not know if the seeds really had the Bt gene in them. The Bt cotton seeds and plants look normal and it was not to separate them from normal seeds or plants. The Central Institute for cotton research Nagpur developed a simple test method like a litmus test paper that could tell the difference and identify the Bt gene in pure Bt cotton seeds within 10 minutes at Rs 10 per test.



The test kits which can be used by farmers directly in the field and also at the shop to check the seed quality on the spot, were commercialized as 'Bt-Express'. In the initial few years, from 2002 to 2005 hundreds of farmers mostly from Gujarat, Maharashtra and Andhra Pradesh used the kits at the sowing time to find out if the seeds they had purchased were genuine. The kits are in use now by farmers and seed testing laboratories across the country.

The Bt-detection kits enabled regulation, streamlining and ensuring Bt-cotton seed quality for farmers in the country. All seed testing laboratories in India have been using the kits and thousands of seed lots have been tested using the kits. Legal cases have been filed in courts of several cotton growing states of north, central and south India and are under review. In the absence of the testing kits, illegal Bt-seed

would have been rampant and proliferated without any control. It has been widely acknowledged that the kits acted as deterrents for spurious seed traders.

90. What is the status of spurious Bt-cotton hybrid seeds in the market?

Data indicate that in 2003 about 69% of Bt cotton brands and seeds sampled from markets were illegal brands and spurious. Current estimates range from 0.5 to 1.0% of the Indian seed market may be under illegal/spurious brand Bt-cotton seeds. Surveys conducted by CICR showed that only 25% of the packets being sold as illegal Bt-cotton are F-1 seeds. About 30% of the packets do not contain any Bt-seeds at all. The rest of 50% is F-2, F-3 or poor quality seed mixtures.

91. What are illegal, fake and spurious seeds?

Bt-cotton was being sold in India for at least two years before it was approved for commercial cultivation. Navbharat-151 was one of the earliest brands of illegal Bt-cotton that was available in Gujarat initially, but later spread to other parts of the country. The cost of illegal Bt-cotton brands was reported to be half of the regular price of Bt-cotton.



Though many farmers found the illegal brands to be profitable, the studies conducted by CICR showed severe quality constraints with many brands of illegal Bt-cotton. Hence, farmers were strictly advised to purchase Bt-cotton seed packets only through proper bill/receipt, so as to avoid falling prey to 'illegal Bt cotton'.

There are also problems of duplicate or fake Bt-seeds being sold as legal and genuine seeds. Wherever there is doubt, the seeds or plant parts may be tested with the Bt-detection kits developed by CICR, Nagpur. The kits are simple to use and can be used directly by farmers themselves. The kits are commercially available with CICR, Nagpur.



92. Are there problems with IPR?

Several genes and components of gene constructs are under patents filed by multinational companies. These genes cannot be used directly in GM crops for commercial purposes. There is an imminent need to discover new genes, promoters from indigenous sources to develop indigenous GM cotton for trait-expression and also gene silencing through RNAi (RNA interference)

93. Can Bt-cotton contribute to food-security?

Profitable cultivation of cotton enhances the purchasing power of farmers and ensures their food security. This is also the case for many countries in Africa where their food security depends on the cotton production.

94. What is the status of public sector cotton in India?

The public sector system of IIT, Kharagpur developed Bt-cotton with Cry1Ac and commercialized through JK seeds in 2006. The National Botanical Research Institute NBRI, Lucknow also succeeded in developing Bt-cotton using Cry1EC. They sold their technology to JK seeds and a few other companies. Currently, there a few Bt-cotton events developed by CICR and UAS Dharwad which are being tested under permission from the RCGM. These may take at least a few years for commercial approval.

95. What kind of policies will help in enhancing cotton yield in India?

The GEAC and RCGM deal with biosafety approvals of events. ICAR should evaluate agronomic suitability and resistance to local biotic and abiotic stress of specific Bt-cotton hybrids or varieties for specific agro-eco-zones. While the approval of new GM-events should be done by the GEAC under the Ministry of environment, GM-varieties and GM hybrids must be approved by the ICAR / SAUs and the Ministry of Agriculture.

Specific productivity enhancing cotton policies should be formulated separately for the 60 districts in India, which cultivate more than 50 thousand hectares of cotton. Twenty of these districts cultivate cotton in 50-90 thousand hectares, another twenty districts cultivate cotton in 91-150 thousand hectares and rest of the twenty districts cultivate 150-430 thousand hectares in each, with a total of 50 lakh hectares.

Each of the 30 Seed Companies should be encouraged to enlist district-wise hybrid suitability of their hybrids and sell only one or maximum two of their best hybrids in a district

Ensure seed purity of hybrids and varieties, especially trait purity in the Bt-hybrid seeds.

Facilitate the introduction of competitive Bt-cotton products from public sector or other technology providers so as to enhance diversity of genes, competitive pricing and enhance sustainability through increased spectrum of genes.

96. Which are the policies and IPM strategies that may augment cotton pest management?

Permit Bt-cotton commercial hybrids only if they are resistant to jassids and major diseases of the zone. About 90% of the current Bt-hybrids are susceptible to jassids and whiteflies. Therefore insecticide usage for sucking pest control in cotton has been increasing over the past five years from 2006 to 2011.

There is a need to introduce new environment-friendly insecticides for sucking pest control. Jassids and whiteflies are showing high level of resistance to almost all the recommended insecticides.

Intensify biological control (inundative and inoculative releases of *Aenasius, Promuscedia* and *Cryptolaemus* spp.) and IPM especially for the emerging pests and diseases, especially for the management of mealy bugs and mirid bugs.

Facilitate dissemination of the Insecticide Resistance Management Strategies for sucking pests and Cry toxin resistance management in bollworms. The strategies should be implemented immediately in Gujarat to delay any possible resistance development.

Mandatory spray on Bt-cotton with Spinosad or biopesticides HaNPV+SINPV at 120-140 DAS may be carried out to reduce the residual resistant bollworm population on Bt-cotton so as to delay bollworm resistance development to Bt-cotton.

Innovate new approaches for planting of refuge by the farmers for sustenance of Bt technology should be developed.

97. Are there any specific policy issues for cost effective profitable cotton cultivation in rainfed regions?

Increase irrigation facilities and drip irrigation in Vidharba region of Maharashtra either through the PM relief fund or Rashtriya Krishi Vikas Yojana.

Encourage pre-monsoon sowing systems wherever possible in Central India. Intensify water harvesting and Integrated Nutrient Management in at least the 20 main cotton districts that cultivate cotton in more than 1.5 lakh hectares.

Intensify extension efforts to educate farmers on the appropriate effects and potential of GM technologies in cotton and biosafety related issues.

Promote high yielding Desi varieties with 18 mm, 16-18 mm g/tex of 6-8 micronaire that are ideally suited for surgical cotton. There is a high demand for such cotton and surgical industry does not have any sources due to decline in Desi cotton cultivation.

Organic cotton can be cultivated in Gujarat and Karnataka using *G. herbaceum* varieties and in Maharashtra with *Gossypium arboreum* varieties in about 5-6 lakh hectares.

98. Is GM labeling necessary?

More than forty countries including the entire European Union, Australia, China, Japan and Russia adopted labeling of GM products as a mandatory requirement. It is widely felt that labeling is necessary to enable consumers make their choice whether or not to use GM foods or products.

The Ministry of Consumer Affairs, Government of India, in an extraordinary gazette notification published on June 5, 2012, has made an amendment to make labeling of every package containing genetically modified food mandatory from January 1, 2013. The notification states that "Every package containing the genetically modified food shall bear at the top of its principal display panel the words 'GM.'

99. Will India lose competitive edge if we slow-down on GM technology?

Depriving farmers of any advanced technologies, such as GM Bt-cotton products will be a retrograde step. Clearly several issues related to Bt-cotton have been with reference to the hybrids and nothing per-se with the Bt-technology itself, it would be improper to brush aside the benefits that the technology has given so far. Considering the fact that cotton is a commercial crop that provides livelihood and purchasing power to the farmer, it would be most appropriate to utilize the best of all available technologies in the most environment friendly manner for profitable cotton cultivation.

In an open market economy under the WTO regime, it is important that the Indian farmer retains competitive edge internationally to be able to use the best available technologies and cultivate cotton at the most competitive costs with high profitability levels. By blocking its doors on advanced technologies it is possible that India will lose its stand in the international markets, and without its own GM technologies, we may have to import agricultural products without having the option of preventing imports of GM goods.

100. Can India become a world leader in cotton?

India has all the potential to emerge as a world leader of cotton. India has the largest cotton area in the world with about 120 lakh hectares accounting for almost one-third of the global cotton area. It has probably the best dedicated scientific talent of the world for cotton research. With carefully planned policy on cotton research we can ensure that the emerging challenges facing cotton farming are addressed from time to time, while harnessing the full potential of our natural resources, manpower and

technologies so that cotton farming becomes a sign of prosperity and India emerges as a global leader of cotton

GM cotton will continue to play an important role. With proper stewardship of approving only better Bt-cotton hybrids that are resistant to sucking pests and other regional biotic stress, coupled with availability of straight Bt-varieties will enable appropriate placement of Bt-cotton hybrids based on their suitability for specific agroeco zones. Bt-Varieties can enhance yields in rainfed regions under high-density planting, while hybrids can harness the potential in other high-input intensive regions of the country, to take India forward to global leadership position in cotton.



References

- Anonymous (2010). A decade of EU-funded GMO research (2001-2010). Directorate-General for Research and Innovation. Biotechnologies, Agriculture, Food. European Union. 2010.p. 16.
- Amman Klaus (2009) Human and Animal Health Rebuttal to a Review of Dona and Arvanitoyannis 2009, part one. *European Federation of Biotechnology*, 2010
- Aumaitre A (2004). "Safety assessment and feeding value for pigs, poultry and ruminant animals of pest protected (Bt) plants and herbicide tolerant (glyphosate, glufosinate) plants: interpretation of experimental results observed worldwide on GM plants". *Italian Journal of Animal Science* **3** (2): 107–121.
- Balachandar, D., P. Raja, K. Nirmala, T. R. Rithyl, S. P. Sundaram (2008). Impact of transgenic Bt-cotton on the diversity of pink-pigmented facultative methylotrophs. World Journal of Microbiology and Biotechnology, 24, 2087-2095.
- Bambawale, O. M., Singh, A., Sharma, O. P., Bhosle, B. B., Lavekar, R. C., Dhandapani, A., Kanwar, V., Tanwar, R. K., Rathod, K. S., Patange, N. R. and Pawar, V. M (2003). Performance of Bt-cotton (MECH-162) under integrated pest management in farmers' participatory field trial in Nanded district, Central India. *Current Science* 86: 1628-1633.
- Craig, Richard (2010) Response to "Health Risks of Genetically Modified Foods" from Dona and Arvanitoyannis (2009) in Critical Reviews in Food Science and Nutrition (49:164-175) *Critical reviews in food science and nutrition*, **50**, 85–91,
- Domingo J. L (2007). "Toxicity studies of genetically modified plants: a review of the published literature". Critical Reviews in Food Science and Nutrition, 47, 721–33.
- Dona, Artemis et al (2009). Health Risks of Genetically Modified Foods. *Critical Reviews in Food Science and Nutrition*, 49,164–175,
- Dong, Liang, Wan Fang-Hao, Zhang Gui-Fen, Liu Xiao-Jing, Li Qiang. (2003). Impacts of transgenic Bt cotton on the development and fecundity of *Chrysopa sinica* Tjeder. *Chinese Journal of Eco-Agriculture*, **11**, 16-18.
- Flachowsky G, Chesson A, Aulrich K (2005). "Animal nutrition with feeds from genetically modified plants". *Archives of Animal Nutrition*, **59**, 1–40.
- Head, G., J. B. Surber, J. A. Watson, J. W. Martin, J. J. Duan. (2002). No detection of Cry1Ac protein in soil after multiple years of transgenic Bt cotton (Bollgard) use. *Environmental Entomology*, **31**, 30-36.
- Hilbeck, A et al. (2012). A controversy re-visited: Is the coccinellid Adalia bipunctata adversely affected by Bt toxins? Environmental Sciences Europe 2012, 24, 10

- Hu, H. Y., X. L. Xiao, W. Z. Zhang, J. G. Sun, Q. W. Zhang, X. Z. Liu, Y. Yu. (2009). Effects of repeated cultivation of transgenic Bt cotton on functional bacterial populations in rhizosphere soil. *World Journal of Microbiology and Biotechnology*, 25, 357-366
- ICAC 1998a Cotton World Statistics, ICAC, Washington
- ICAC 1998b Survey of the costs of production of raw cotton. ICAC, Washington
- ICAC-CCRI 1999. Proceedings of the Regional Consultation on Insecticide Resistance Management in Cotton. 28 June – I July 1999. pp 293. Central Cotton Research Institute, Multan, Pakistan
- Kapur, M., R. Bhatia, G. Pandey, J. Pandey, D. Paul, R. K. Jain. (2010). A Case Study for Assessment of Microbial Community Dynamics in Genetically Modified Bt Cotton Crop Fields. *Current Microbiology*, **61**, 118-124
- Kathage J and Qaim M. (2012) Economic impacts and impact dynamics of Bt (*Bacillus thuringiensis*) cotton in India, *PNAS*, **109**, 11652-11656.
- Key S, Ma JK and Drake PM (2008). "Genetically modified plants and human health". J R Soc Med 101, 290–8
- Kranthi, K. R., Russell, D., Wanjari, R., Kherde, M., Munje, S., Lavhe, N. and Armes, N. (2002). In-season changes in resistance to insecticides in *Helicoverpa armigera* (Lepidoptera: Noctuidae) in India. *Journal of Economic Entomology*. **95**: 134-142.
- Le Curieux-Belfond O., Vandelac L., Caron J., Seralini G.E. (2009) "Factors to consider before production and commercialization of aquatic genetically modified organisms: the case of transgenic salmon" *Env. Sci. Policy* **12**, 170–189
- Lu, Y., Wu, K., Jiang, Y., Guo, Y., Desneux, N (2012). "Widespread adoption of Bt cotton and insecticide decrease promotes biocontrol services". *Nature*. doi: 10.1038/nature11153.
- Magaña-Gómez J. A, de la Barca A. M (2009). "Risk assessment of genetically modified crops for nutrition and health". *Nutr. Rev.* 67, 1–16.
- Mayee, C. D (1997) Parawilt of cotton: nuances and the challenges. J. Mycol. Plant Pathol., 27, 28
- Netherwood et al. (2004). "Assessing the survival of transgenic plant DNA in the human gastrointestinal tract". *Nature Biotechnology*, **22**, 204–209.
- Rui,Y. K., G. X. Yi, J. Zhao, B. M. Wang, J. H. Li, Z. X. Zhai, Z. He, Q. X. Li. (2005). Changes of Bt Toxin in the Rhizosphere of Transgenic Bt Cotton and its Influence on Soil Functional Bacteria. *World Journal of Microbiology and Biotechnology*, 21, 1279-1284.

- Sarkar, A. K. Patra, T. J. Purakayastha, M. Megharaj (2009). Assessment of biological and biochemical indicators in soil under transgenic Bt and non-Bt cotton crop in a sub-tropical environment. *Environmental Monitoring and Assessment*, **156**, 595-604.
- Séralini et al. (2011) "Genetically modified crops safety assessments: present limits and possible improvements" *Environmental Sciences Europe*, **23**, 10
- Seralini G.E., Cellier D., Spiroux, de Vendomois J (2007) "New analysis of a rat feeding study with a genetically modified maize reveals signs of hepatorenal toxicity" *Arch. Contamin. Environ. Toxicol* **52**, 96–102
- Séralini, G. E., et al. (2012) Long term toxicity of a Roundup herbicide and a Roundup-tolerant genetically modified maize. *Food Chem. Toxicol.*, <u>http://dx.doi.org/10.1016/j.fct.2012.08.005</u>
- Snell, C, Bernheim, A, Bergé, JB, Kuntz, M, Pascal, G, Paris, A, Ricroch, AE. (2012) Assessment of the Health Impact of GM Plant Diets in Long-Term and Multigenerational Animal Feeding Trials: a Literature Review. *Food and Chemical Toxicology*, **50**, 1134–1148
- Udikeri, S. S., (2006), Evaluation of new generation Bt cotton genotypes, Sustainability of Cry protein expression, computation of ETL, Effect on aphid predators and development of IPM module for Bt Cotton under rainfed conditions. Ph.D. Thesis, Univ. Agric. Sci., Dharwad, Karnataka, India
- Udikeri, S. S., Patil, S. B., Nadaf, A. M. and Khadi, B. M (2003). Performance of Bt-cotton genotypes under unprotected conditions. *Proceedings World Cotton Research Conference-3*. Swanepoel, A. (Ed) Agricultural Research Council-IIC, Cape town, South Africa. 2003: 1282-1286.
- Vain, P (2007). "Trends in GM crop, food and feed safety literature". Nature Biotechnology, 25, 624.

About the Author



Dr K. R. Kranthi is currently the Director of the Central Institute for Cotton Research Nagpur. His research interests are focused on the utilization of biochemistry and molecular biology for the development of simple farmer usable technologies for sustainable pest management. Dr Kranthi has been working on cotton at the CICR from 1991 and has developed several diagnostic kits to detect Biotech cotton, insecticide resistant insects and substandard insecticides.

The kits were commercialized and are being used by all the seed testing agencies, farmers and the seed industry and have directly contributed to the reduction in spurious Bt-cotton seed in India. He worked on the biochemistry, genetics and stochastic modeling of bollworm resistance to insecticides and biotech cotton. Several of the insect resistance management strategies developed by his group have been approved by the Government of India. The IRM strategies are being successfully used by more than 50,000 cotton cultivators across India over 10 years for effective pest control through 40-60% less pesticides.

A gold medalist in Ph.D from the Indian Agricultural Research Institute, New Delhi, Dr Kranthi has two patents in India and four in South Africa, South Korea, China and Uzbekistan. He has published 35 papers in peer reviewed journals. He was elected as the fellow of the prestigious National Academy of Agricultural Sciences, India in 2009. Dr Kranthi received four National Awards in India and was recently recognized as the 'International Cotton Researcher of the Year Award 2009' by the International Cotton Advisory Committee, Washington and was felicitated at Bremen, Germany by the cotton fraternity in 2010 for his research contributions.